

Rules and Regulations for Test-tube Babies

ALTHOUGH the authorities in Britain which regulate the pedigrees of racehorses, not to mention the Kennel Club and the smaller societies which attempt to prevent miscegenation among breeds of dogs and cats, still set their faces against artificial insemination for pedigree blood-stock, there is now no such restraint where people are concerned. Interference with the natural course of human reproduction has gone much further. Abortion is now legal in many countries, on social as well as eugenic grounds. Some genetic defects can be recognized sufficiently far before birth for prophylactic abortion to be safe. Most probably, sex determination is round the corner. Dr R. G. Edwards and his colleagues at Cambridge have for some time been inspired by the notion that it may be possible to avoid some kinds of infertility in women by the artificial fertilization of an ovum and its transfer to the uterus of the donor after a few days of development.

By now, a great many experiments with laboratory animals suggest that artificial fertilization is only one of the many techniques which may in the course of time make it possible to regulate more precisely than is at present possible the outcome of the reproductive process. The article by Dr Yu-Chih Hsu on page 100 shows how (with mice) laboratory work is helping to span the gaps which at present remain in the artificial development of complete embryos—the development of an artificial uterus that will allow a mouse blastocyst to develop to the point at which its heartbeat shows is a considerable achievement. The experiments with amphibian eggs by means of which Dr J. D. Gurdon and his colleagues at Oxford have been able to grow several identical organisms from a single somatic cell in much the same way as the botanists have been cloning carrots from single cells have excited a great deal of curiosity among the prophets of artificial reproduction. Will it one day be possible to use similar cloning techniques to produce identical copies of selected individuals, Albert Einstein perhaps or Rudolph Valentino? It is no wonder that questions have been asked about the propriety of research like this and about the means which should be adopted for guiding it in beneficent directions. On page 87, Dr R. G. Edwards and Dr David J. Sharpe provide a definition of some of the matters which cry out for attention and put forward some practical suggestions which cannot be ignored.

What are the dangers? And what are the benefits? The first need is to acknowledge that the physiology of reproduction is an important field of research from which great advantages may derive. More effective (which usually means simpler) methods of contraception would be a great boon. But reproductive physiology is also potentially important for the understanding which it may provide of the processes of differentiation in developing organisms, the mechanisms by which genetic defects are made apparent and even the nature of intra-uterine life. Then there are great prizes to be won even in some of the far-fetched schemes for interfering with the natural course of reproduction which are often trotted out among the list of the potential horrors of the modern world. There would, for example, be immense economic benefits in

schemes for determining in advance the sexes of domestic livestock; dairy farmers are at present required to forfeit half the reproductive potential of their herds because bull calves are worth no more than their weight in meat. Then what if it were practicable to endow cells with genetic properties derived from other strains by the mere transplantation of chromosomes, thus improving the productive potential of domestic animals? Is there an agriculturalist who would desist? Even cloning, which provokes awesome and unfamiliar questions, would no doubt be a great advantage if it were a method of making sure that animal herds were uniform and excellent in composition. To be sure, there might be unexpected if second-order difficulties, but in principle there is nothing in even the most outrageous of the possibilities which Edwards and Sharpe enumerate to contradict the traditional view of animal breeders that devotion to the genetic improvement of stocks of animals is a virtue. What this implies is that the potential benefit of artificial reproduction in animal breeding would by itself be a sufficient justification of the laboratory experiments now being undertaken.

The difficulties multiply where people are concerned. First of all, the benefits are far less obvious. Dr Edwards is right to argue that much human discontent would be removed if there were a way of avoiding those kinds of infertility which can be treated by artificial fertilization followed by implantation, but in an overcrowded world infertility is an individual problem, not a great social issue. Similarly, there are serious philosophical doubts about the goals to which human eugenicists should strive. It is easy enough to say that the best racehorses are the fastest, but, with people, there are difficulties in knowing whether apparently desirable characteristics such as intelligence are entities in their own right or, alternatively, products of the society to which they contribute. Worse still, for the would-be genetic engineers, it is more than likely that all the techniques of artificial fertilization, artificial embryogenesis, artificial cloning and chromosomal translocation are subject to errors that may be acceptable where animals are concerned but which would almost certainly among people do more harm than good. Which putative parent, after all, would set out to produce children by cloning if there were even a remote chance that the incidence of known genetic defects, trisomic mongolism, for example, were greater than in natural reproduction? Even with the large manipulable amphibian eggs used so far in cloning experiments with animals, defects of reproduction are common. There is a high chance that all the proposals so far put forward for manipulating human embryos in the course of development will entail so much risk that they will not be lightly used. In other words, when applied to human beings, artificial reproduction and genetic engineering will always be required to contend with a serious and unavoidable natural impediment.

So is there no need for anxiety? Can the natural restraints of modern society be reckoned as sufficient to keep experiment in check? Drs Edwards and Sharpe are right to say that the problem is larger than this. To begin with, there is bound to be experiment with human as



well as animal material. Dr Edwards's own blastocysts have on many occasions been washed down the laboratory sink. There is nothing to prevent laboratories elsewhere from carrying out attempts at the cloning of human cells, and there is at least a remote chance that some of them may succeed. Certainly there will be attempts, entirely well intended, to see whether it is possible successfully to mask genetic defects by infiltrating into an embryo at a very early stage of its development a cell which can make up for the genetic defect of its host. This, in the long run, is probably a simpler route to genetic engineering than chromosomal transplantation. The trouble, of course, is that no experiments like these can be undertaken without raising doubts in the minds of those not fully aware of what is going on about the outcome and the significance of research like this. The chance that some reproductive physiologist may one of these days find his laboratory peopled by a clone of identical human beings is fantasy, not a real danger; the fear should be that the social significance of the discoveries yet to be made may be overlooked by those primarily responsible and misunderstood by those outside the field.

In circumstances like these, is there not a strong case for asking that among reproductive physiologists there should be a much fuller exchange of information than is usually the case? The first objective, and the minimum, should be the provision of a professional forum in which scientists can obtain either advice or courage in the conduct of their work. Here, as everywhere, the chances are high that problems which seem overwhelming to individuals have already been dealt with by other workers in the field. But is there not also a case for asking that this exchange of information should be monitored by people outside reproductive physiology as such? In spite of the apparent importance of legal and even philo-

sophical questions in discussions like these, to be sure, it is not immediately apparent that the ideal forum for these discussions should be as broad as that which Edwards and Sharpe define. On the face of things, a good beginning could be made within the framework of academic science and medicine. The objective should be a publicly known body of professionals, internationally based, which would be able by its reputation to command the respect of scientists working in this important field and, by doing so, would be able to ensure not merely that most innovations became quickly known but also that most innovators were constrained by the pressures of their professional loyalty to talk problems out.

Nobody could guess at this stage whether such a body would wither away, if it turned out to be less useful than seems probable, or whether it would become a pragmatic institution devoted to the ethical problems of reproductive physiology. Drs Edwards and Sharpe mention the first possibility, but it is more likely that the future will only serve to strengthen the need for some formal means by which scientists can make a clean breast to fellow professionals of their objectives in embarking on novel fields of research. Moreover, the risks of misunderstanding being what they are, it is almost certain that only by being seen to have some mechanism for registering objectives will reproductive physiologists be able to avoid unreasoning public criticism. There is even a chance that a properly constituted and sufficiently authoritative body of opinion in this field could so help to clarify the problems which at present lurk at the backs of people's minds that research could be accelerated and not held back. That, in all the circumstances, would be a sufficient excuse for setting up the right invigilatory commission. Is this not a task to which the learned academies, particularly in Britain and the United States, should lend their weight?

No Dole at the Top

THE losing team in the gladiatorial football match with which the British football season ended last week has been for the past twelve months singled out by all writers on the subject for having a university graduate on its roster. By all accounts, Mr Michael Heighway, who plays for Liverpool, is the only professional footballer in Britain to have been to university and to have come away with a degree. For professional football, no doubt, this remarkable statistic will in due course raise important problems. For how long will a successful sport be able to live off the talents of those who are academically less able than those who find their way to universities? At the worst, it might have been expected that people's talents in scholarship and at football were entirely independent variables. Given the small but significant correlation between intelligence and physical skill that psychologists have established, and the apparent capacity of football clubs to pay very large sums of money for recruits, is there not a case for thinking that prudent football managers should flourish their cheque books under the noses of many more university graduates? Certainly as far as the universities are concerned, such a development would be entirely welcome. For a year now, British universities have been echoing the earlier fears of their American counterparts about the difficulties of finding jobs for graduates. A week ago, the House of Commons

engaged the handful of its members still left on Friday afternoon in a brief and nostalgic debate on the subject. It is clear that there is some way to go before British legislators and British taxpayers are reconciled to all the implications of what has become nearly universal higher education.

For Britain, the plain truth is that the success of universities and other institutions of higher education in growing to meet the demands of students and parents for higher education has saturated the previously existing labour market. The signs of strain have been apparent in the months since last year's crop of 60,000 graduates returned to Britain from their summer holidays and began looking for jobs. Quite soon, there will be another 60,000 graduates on the labour market and nobody can be cheerful about the prospect. One estimate is that 2,500 of last year's crop were still looking for work by the end of 1970, and it is more than likely that a still larger number of unemployed graduates had fallen through the statistical network of the Universities Appointments Board. In circumstances like these, it is easy enough to jump to all kinds of incorrect conclusions. In the House of Commons, for example, Mr Mark Hughes, who raised the complaint about unemployed graduates with the government, asked that there should be a reappraisal of the role of the universities in British social life. In par-

ticular, he asked that "universities must accept that they have a role to provide their students with a bread ticket". The idea is that universities should be less like "ivory towers" and more concerned with the careers of their graduates. Because of the danger that Mr Hughes's message will become the popular interpretation of what seems to be a temporary surplus of graduates, and because this may in turn lead to a misapprehension of what universities are for, it is important that his heresy should be recognized for what it is.

To begin with, it is difficult to be overwhelmed by the plight of 2,500 graduates when 800,000 other people, the vast proportion of them with family responsibilities, are unemployed in Britain. Indeed, when it appears that more than three per cent of the entire British work force is out of work, graduates with a strong sense of proportion may think it lucky that the proportion of last year's graduates still unable to find a suitable job is no greater. Evidently graduates are still more able than unskilled workers to find a niche for themselves in the British economy. The difficulty, given the economic circumstances of the present, is therefore not the immediate problem of unemployment but the question whether there is now likely to be a permanent imbalance between the output of the universities and the demand for labour of the traditional employers of university graduates. It is only fair to acknowledge that in present circumstances the truth must be exceedingly obscure. It is well known that in times of economic difficulty, the mobility of professional people is diminished. Where graduates are concerned, for example, what this implies is that there are fewer vacant places in professions such as the law or chemical engineering because fewer lawyers have spread their wings into other professions or fewer chemical engineers have found themselves snatched away to become tycoons. This shortage of openings could easily be a temporary recession, and could be followed in a year or so by as clamorous a demand for graduates as any to have gladdened the hearts of appointments officers in the past few years. In other words, a part of the present depression of the graduate labour market is a marginal phenomenon, not a permanency.

It is also likely, however, that some part of the present difficulty of finding jobs for graduates is linked with the way in which the scale of higher education in Britain has grown in the past two decades more rapidly than the national economy. Since the early fifties, university education in particular, and higher education as a whole, has been growing at such a pace that the output of the universities has multiplied by four in two decades. It is beyond belief that such an expansion could be accomplished without qualitative changes in the character of the labour market. In particular, it is no longer reasonable to expect that the proportions of graduates ending up as teachers should be as great as in the forties or fifties. Indeed, a large part of the incentive for increasing the whole scale of the operation has been the desire to broaden the uses to which advanced education could be put. For decades, British industrialists have been saying that the British economy would be only thoroughly competitive with, say, the German economy when it became possible for technicians in British factories, like those elsewhere, to be proud of having some kind of degree. This, of course, is the case of Mr Heighway. The plain truth is that in terms of the labour market, what is wrong with the output of graduates from the universities in

Britain is not the number of educated people being produced but the conservatism that supposes that higher education in the 1970s must serve exactly the purposes to which higher education in the thirties was put.

What are the implications for the universities? Mr Mark Hughes was, of course, entirely wrong in suggesting that when the need for a greater diversity of occupations for graduates should be encouraged, universities themselves should seek to provide their graduates with training that enables them more effectively to compete for particular kinds of jobs. The argument should be turned the other way, and when it is increasingly necessary for graduates to look for diverse occupations, universities should be more anxious than ever to train them to be flexible. Paradoxically, in other words, the new circumstances which have caused such consternation among universities and their graduates should be regarded not as a spur to more specialized education but the opposite, yet another indication that British higher education has become too detached from the old-fashioned belief that education is worth having for its own sake.

100 Years Ago



THE PEOPLE'S UNIVERSITY

A GIGANTIC and imposing educational scheme is about to be launched, which, whether it proves feasible or not, must attract the attention and enlist the sympathy of all well-wishers to the intellectual development and material welfare of the country. This is no less an idea than the establishment of a National Working Men's University, which is to be founded with special reference to instruction in those subjects which have a direct bearing on the arts and manufactures. That our workmen are, as a rule, altogether ignorant of the scientific principles upon which the processes they ought to guide and govern are dependent, and that England in this respect stands in a much inferior position to continental nations, is now a well-recognised fact. The result of this lamentable ignorance is stated by certain authorities to be severely felt in those of our trades and manufactures in which we have to compete with other nations; and although this conclusion has been denied by many, yet concerning the necessity for scientific education amongst our artisans there has never been a difference of opinion. The question then arises, How are we to bring to our rising artisans on an extended and national scale the knowledge of scientific principles which they so much need, and for which the best of their class show so much desire and even aptitude? One solution to this problem is being attempted by the scheme of a National University for Industrial and Technical Training. The proposal is to establish a metropolitan institution in which complete and thorough instruction in all those branches of knowledge which are of importance to our manufacturing industry shall be given.

From Nature, 4, 41, May 18, 1871.

OLD WORLD

PASTEUR INSTITUTE

Monod's New Parish

Paris, May 11

THE Pasteur Institute seems not yet to have escaped from its difficulties of the past few years. For some time, it has been short of money, racked by internal conflict, while its enterprise in manufacturing vaccines has been increasingly coveted by private industry. The appointment of a new board of directors has helped to take off some of the strain, and the appointment of Mr Jacques Monod, the Nobel prize-winner, as the new director has been a great encouragement to the institute. But now, as luck will have it, the institute finds itself threatened by another crisis, the difficulty of finding room in which to grow.

The issue was brought to a head on May 5 at a public meeting called by the Comité Central d'Entreprise, the constitutional body which is elected by employees of the Pasteur Institute. The issue that now disturbs the institute stems from a comparatively simple administrative problem. Although most of the research has traditionally been carried out in central Paris, it seems to be acknowledged that the Pasteur Institute needs extra space for its production plants and for some time it has been planning to erect several new buildings on a 100 hectare site at Rennemoulins, in the suburbs.

This proposal has fallen foul of the French Government's regional planning unit, the champion of planned dispersal from Paris and its suburbs, which argues that the Pasteur Institute should put its production facilities at Louviers in southern Normandy. The institute complains that this town is a long way from airports and railheads and a thoroughly inconvenient place from which to ship vaccines to the four corners of the Earth when there are epidemics. The institute is, moreover, disinclined to spend roughly Fr.8 million (about £600,000) on a site and to train the staff needed to replace those of its present employees who would decline to move 75 kilometres west.

The Pasteur Institute has won support in its struggle to stay near Paris from the Ministry of Social Security and Public Health, the Opposition in Parliament and a substantial part of the Government party. It has now appealed directly to the President, Mr Georges Pompidou, and to the Prime Minister, Mr Jean Chaban-Delmas. Then, making the fullest use of its reputation, the institute is appealing directly to the public.

The latest crisis has been building up

while Mr Jacques Monod has been trying to concentrate on the improvement of the institute's long-term financial prospects, and particularly on the possibility of recovering some of the ground lost to private industry in the past few years in the manufacture of pharmaceutical materials. The immediate objective seems to be the development of new products, the working out of a more aggressive commercial strategy and the strengthening of the institute's several research departments, especially so as to avoid duplication. There is also the prospect that the institute might increase the scale of its business, on the success of which the survival of the research departments depends. It is also hoped that it will be possible to arrange licence exchange agreements with the Wellcome Foundation in the United Kingdom and the Rockefeller Foundation in the United States.

SOVIET RESEARCH

Where to Push Next

from our Soviet Correspondent

THE directives for the new five-year plan of the Soviet economy, published last February, contained a broad scheme of basic and applied research projects for the next five years. Unfortunately, the proposed outlines are so all-embracing, and the fields of research so diverse—from cybernetics to geochemistry and from semiconductors to the genetics of hereditary diseases—that it has proved virtually impossible to conclude from the directives what will be the chief lines of Soviet research in the next half-decade. Nor did the speech of Academician Mstyslav V. Keldysh, President of the Academy of Sciences, to the Twenty-Fourth Congress of the CPSU, greatly amplify the position, concerned as it was chiefly with a review of past achievements, and general hopes for future successes. Perhaps the most concrete indication of policy which could be drawn from his speech (see *Pravda*, No. 92, 1971) was his emphasis on the serious deficit in the production of scientific instruments and the need for an acceleration of output in this field, with the implication that unless a considerable improvement is obtained, this lack could well bedevil the whole plan for the expansion of science and technology.

Now, however, addressing the May meeting of the Academy of Sciences, Academician Keldysh has given some clear indications of which research projects will bear greatest emphasis within the context of the current five-year plan. According to Soviet planning theory, all research is to be closely linked with

its ultimate practical application. "In planning the development of science, we must constantly have in view that the functions of science in the development of contemporary society are exceptionally broad and a manifold complex of questions . . . rests on the whole system of the social and natural sciences."

More specifically, in implementing the general directives, special emphasis is to be placed on cybernetics in all its ramifications—information processing, the technology of automation and control and the development of computers, especially for planning purposes. Research in physics is also to be emphasized, with special attention to solid state physics, electronics, radio-physics and nuclear research. Fuller use is to be made of existing high-energy accelerators and the experimental basis of medium-energy nuclear physics is to be "strengthened". Plans for space research (Keldysh's own particular field of interest) are fairly vague, but the significance of the *Salyut* project is stressed, and plans for further study of the Moon and planets by automatic spacecraft are indicated. (There is no mention of manned missions beyond Earth-orbit.)

In general, the implementation of the directives seems to fall chiefly upon physical and mathematical research, save for the emphasis, yet again, on the need for more "rational" use of natural resources. Russian textbooks traditionally describe the Soviet Union as the "richest country in the world in natural resources", and under earlier plans, it was tacitly assumed that these resources were inexhaustible. The past few years have, however, shown a considerable reversal of this attitude, and Academician Keldysh, in his address to the academy, reiterated the five-year plan directives yet again in emphasizing the attention to be paid to study of the mineral, agricultural and water resources of the Union as the basis for further economic planning and the development of the country.

NUCLEAR POWER

Dangers of Proliferation

CONCERN for the spread of gas centrifuges brought together thirty-four people in London on May 7 to discuss their fears under the auspices of the J. D. Bernal Peace Library and the British Society for Social Responsibility in Science. They were worried chiefly about the consequences for non-proliferation if ultracentrifugal methods of producing enriched uranium, which have been under development since 1946, are widely adopted. One of the

causes for concern is the agreement, completed last year, between Britain, the Netherlands and West Germany, which marked the start of a collaborative development programme, adding to the work already in progress in the United States, the USSR and other countries.

Dr Colin Sweet, of the J. D. Bernal Library, spoke for all the participants when he said that the primary objective of this project seemed to be economic gain from sales of gas centrifuges. All agreed that before this comes about there must be stringent safeguards which, said Dr A. McKnight of Sussex University, must be of a far higher standard than normal commercial practice. The danger that enriched uranium could be stolen and used to make weapons in secret must be avoided. Dr McKnight was concerned, however, that so far there has been insufficient emphasis on safeguards in the planning of centrifuge plants. Sir Michael Wright of the United Nations Association, a seasoned negotiator on matters of disarmament, pointed out the difficulty of getting agreement on safeguards with the countries of eastern Europe, although he thought it should be possible.

International controls were called for, and Dr Frank Barnaby, a journalist specializing in military technology, suggested that the International Atomic Energy Agency should become a supplier of enriched uranium. He thought the agency could be expanded to become responsible for the installation and operation of all imported nuclear facilities. This and the other fruits of the day's deliberations should be available for all to see when they are published. There seem to be no plans to make the views expressed any more widely heard.

ELECTRICAL ENGINEERS

100 Years in London

THE Institution of Electrical Engineers celebrates its centenary next week with three days of talk and festivity, as befits the largest learned society in Britain. The celebrations begin on Monday with a service of thanksgiving at Westminster Abbey to be followed by the opening ceremony at the Royal Festival Hall. The professional needs of the occasion will be served by a congress on Tuesday and Wednesday.

The institution can look back on a century of success, both in terms of growth and influence. The membership now amounts to more than 63,000, both in Britain and abroad, and thus dwarfs comparable organizations such as the Institute of Physics (15,000) and the Chemical Society (16,500). The

institution is also well-heeled. Its annual report, to be published later this month, will show that the income for the year ended December 31, 1970, was £1,462,360 for an expenditure of only £1,231,510—an excess of nearly a quarter of a million pounds. This income is derived from five chief sources—an extensive list of successful publications, membership fees (£6.30 annually), rents and conference fees and the proceeds of INSPEC, the institution's information service.

The institution owns its own building in Savoy Place, but this can no longer house all of its activities; the publishing department has been banished to Stevenage, and most of the INSPEC staff are at Hitchin.

Through its widespread membership, the institution maintains strong contacts with industry, but, perhaps surprisingly, it seems that industrial finance plays little part in the institution's success. Nevertheless the benefits to be gained from cooperative interchange must be great. Next week may be an occasion for the institution to look back on the past with satisfaction, but it is also a time for looking to the future. And the title of the first lecture in the congress, to be given by Sir Brian Flowers, chairman of the Science Research Council, is "The Scientific Basis of Electrical Engineering in A.D. 2000".

SOCIETIES

Shortage of Homes

WITH the proceeds of its highly successful appeal, the Linnean Society has given its headquarters a new look, 111 years after moving into Burlington House in London. What it seems to need in the future are some new policies to keep it alive now that the naturalists who used to fill its ranks have largely given way to specialist biologists who are not inclined to join a society still devoted to evolution and classification.

Since the appeal for redevelopment was launched in 1969, almost £77,000 has been raised. The most important item of redevelopment has been the strongroom to hold the collections and library of the eighteenth century Swedish botanist Carl Linnaeus. These were acquired by J. E. Smith, a rich amateur naturalist, in 1783, five years before the founding of the society, which has held the collection since Smith's death.

The redevelopment has also included the expansion of the library, the provision of offices for the executive secretary and a council room, as well as redecoration throughout. But more needs to be done, especially to improve the facilities for book storage, and

another appeal has been launched to raise £2,500 to match a similar sum from the Bentham-Moxon Trust. Another much needed improvement, which is expected to be paid for by £6,000 from the Pilgrim Trust, is the reclassification of the library. The society has an important collection of historical volumes as well as an up to date library, but has at present only a catalogue of authors.

Remarkable as all this may seem in a climate of financial restriction, the society is suffering as much as other learned societies. Although the rooms in Burlington House are provided rent free by the government, the cost of upkeep is a heavy burden and the society is not attracting the new members that it desperately needs. Today there are 1,100 members, which is a poor improvement on the mid-nineteenth century when there were already more than 800.

The Linnean Society is at least in a happier position than many learned societies which either have no accommodation or are about to lose it. The greatest need of a society, however small, is for centrally located office space and a committee room where policies can be formulated. The Linnean Society has for many years helped to alleviate this need by making its rooms available to the Malacological and Royal Microscopical Societies and several others. The Institute of Biology, which is rapidly outgrowing its headquarters, provides secretarial staff for fifteen of its member societies, and has a small meeting room which holds 100 people. Few biological societies are as well endowed as the Biochemical Society, which has its own premises in London.

The Society for General Microbiology has bought a house in Reading, much cheaper than equivalent accommodation in London, and as near to the capital as the society wishes. The house provides offices and room for small meetings, but council meetings are still held in London, at the Ciba Foundation.

Other societies regard the idea of moving out of London as a last resort. The Royal Statistical Society, for example, has accommodation in London, but the lease will soon expire. The society is anxious to stay in London, which is so convenient for council meetings and for its library. Considerations of this sort have prompted this and several other scientific societies to show an interest in a six year old scheme to build offices and meeting rooms on a site near Regent's Park. London International Centre Ltd has outline planning permission from Camden Borough Council, which requires the provision of student accommodation together with the office buildings. The difficulty of raising the

money for this student accommodation is now blocking the scheme, although the people involved are less downcast about the prospects than they might be.

As well as providing space for some scientific organization, this centre is intended to house several international voluntary organizations, including the Save the Children Fund. The organizations would buy their own leases, and gain the use of an assembly room, committee rooms and space for the libraries which some would bring with them. London International Centre would solve the accommodation problem for the societies which it housed, but the plight of many others must remain precarious as they try to hang on to expensive London homes, or continue to suffer inadequate or non-existent premises.

CONSERVATION

Protecting Predators

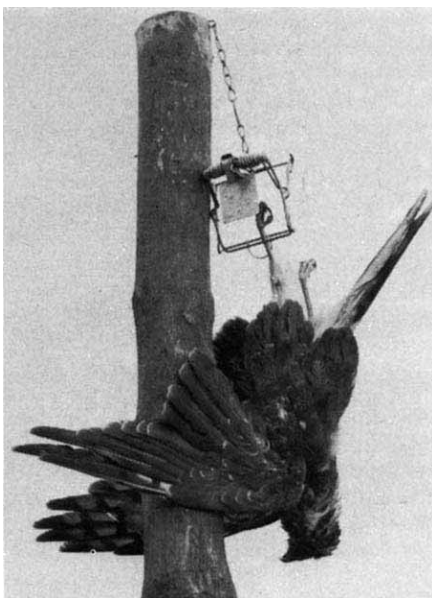
THE Royal Society for the Protection of Birds has launched a campaign to stop the killing of birds of prey. These birds "enjoy" full protection under the law as it stands, yet every year many cases of birds shot, pole-trapped or wantonly poisoned are brought to the notice of ornithologists throughout Britain. This year alone, thirty-one cases are already documented in the RSPB's files and in all but two cases the birds themselves were the intended victims of the so-called control measures.

Virtually all the blame must fall on British gamekeepers. Although fully aware that they are breaking the law, they still operate according to the old prejudice "if it's got a hooked beak, kill it"—a doctrine to which they slavishly adhere in spite of the paucity of evidence to show that game-bird breeding is adversely affected by avian predators. Indeed, most ecological studies indicate that birds of prey serve more to safeguard the breeding success of pheasants and partridges, for example, by destroying small rodents, whose egg-sucking habits are well known. In many cases, particularly among owls, there is no evidence whatsoever of conflicting interests with game birds.

The setting of spring traps in any position in which they are likely to catch birds (this clearly applies to pole traps and open-sited gin traps) has been illegal in Britain for several years; the use of poisoned bait is also against the law except under licence, and then only for certain types of poison applied in a restricted number of ways. Few, if any, such licences have been issued against birds in recent years. Why then is the use of such techniques so blatant and so widespread?

Many gamekeepers, it is certain, hide

behind the protective cloaks of their employers, the landowners (often titled) and wealthy shooting syndicates whose sporting interests they claim conscientiously yet illegally to protect. It is more than probable that many prosecutions have been blocked at source by chief constables anxious to preserve good relations with local dignitaries who may themselves be the magistrates presiding over the courts which hear the cases. Lack of evidence is the most frequent excuse for abandoning prosecutions, and under the law as it stands this is all too easily upheld, for only the person responsible for setting the trap, pulling the trigger or laying the poison can be brought to court. Under these restrictions, only a confession from the offender or the adequate witnessing of the moment that he commits the offence can uphold a prosecution. It is certain that if landowners were held responsible for the management practices conducted on their property, pole traps and poisoning would cease to be a problem.



Hen harrier (*Circus cyaneus*) illegally killed in a gamekeeper's pole trap.

There are other shortcomings of the law. A recent case in Kent outlines just how pressing is the need to give the extensive but surprisingly ineffective legislation some teeth, and also to require magistrates to treat cases with less flippancy. A Rochester gamekeeper admitted to using pole traps and was charged under the Pest Act. The defending solicitor submitted that a bird was not an animal and therefore the case did not fall within the terms of reference of the act. After consultation with the clerk, the magistrate conceded the point and the charge was quashed. On a second charge, brought under the Bird Protection Act, the gamekeeper admitted shooting owls. He claimed that the birds were killing

young pheasants and was asked whether he had any evidence of this (the act condones killing under such circumstances). His reply was no, but the trouble had stopped after the owls were shot. With this glib justification, too easy to give and too frequently used to dodge the law, the man was acquitted.

As well as providing more effective means of administering the law, educating gamekeepers to a better appreciation of the role of birds of prey in the ecological scheme of things might help. It is here that support from ornithological ethologists and ecologists is urgently needed. One of the problems faced by the compilers of a booklet to be published by the RSPB later this year has been the scarcity of studies to which they have been able to refer.

Called *Predatory Birds in Britain* and containing contributions from the Gamekeepers Association, the Game Conservancy and the British Field Sports Society, the booklet aims to offer constructive suggestions on how to handle the few authentically difficult situations which it admits do sometimes arise. Without killing the offenders, it maintains, it is possible to safeguard the interests of both the gamekeepers and the conservators.

It seems that more research, more publicity and modifications of the obviously inadequate protection laws would do much to combat this unnecessary threat to the conservation of a section of the country's wildlife which is already labouring under a heavy burden of habitat destruction, human interference and environmental pollution.

COMPUTER SOCIETY

Looking for Members

THE British Computer Society has launched a concerted effort to double its membership to about 30,000 in the next two or three years, and in this way to consolidate and strengthen its position as an effective central body in the computing profession. Much of the campaign is aimed at existing members of the society, who are being encouraged to introduce prospective members to the society.

The society was founded in 1957 and brought together many informal groups with wide interests in scientific and commercial computing. This mixture is still a characteristic of the society, which now has about forty specialist groups dealing with topics as diverse as the social implications of computing, computer-aided design and auditing by computer. Apart from incidental publications, the society regularly publishes two periodicals—the *Computer Journal*, which contains original contributions, and the *Com-*

puter Bulletin, which is the chief link between the society and its members.

There was a considerable expansion in the membership—from about 3,000 to about 18,000—between 1965 and 1968, but since then the number of members has decreased to about 16,000. The reason for this drop lies in the introduction of a two-part examination system, which required that candidates for membership after 1968 should either pass the examinations or be exempted from them by virtue of other equivalent qualifications such as a degree in computing science, or (at a lower level) a Higher National Diploma in an appropriate subject. This was all part of the society's policy of increasing professionalism in the computing field, and also during 1968 the society began to look seriously at proposals for the computing code of practice which it has published recently.

Predictably there was a rush of applications at the end of 1968 for membership on the old basis of proven experience in computing rather than the new one of paper qualifications, and since then numbers have tended to fall off, chiefly because of wastage, as the examination system became established. Ninety candidates sat the first examinations in 1969, and by April 1971 this number had risen to about 300, some of whom were in Durban, Hong Kong and Singapore.

The society has recently included a clause in its membership regulations which allows older computer practitioners who did not join before 1968 to qualify for membership provided they have at least ten years of proven experience—but no entries will be allowed in this way after 1975.

Financially, the society is in a healthy position, although the president, Mr Alex d'Agapeyeff, has made it clear that if more money were available there would be greater scope for the improvement and widening of the society's activities. He was clearly thinking not only of the extra income from an increased membership, but of the possibility that the government would eventually give the society some form of grant to help it with the work it does in the public interest, and which it has at present to finance from its own funds. The society's own committee on privacy is, for example, collaborating with the office of the Registrar-General in an effort to make absolutely certain that the computing processes involved in the recent census leave as little scope as possible for abuse. During 1969–70 the society had an income from membership subscriptions and conferences of about £135,000, and was able to increase its reserves by £8,950 to £57,742.

Looking to the future, the society is anxious to expand its role as an adviser

on standards and career prospects to educational establishments in the UK and, possibly, throughout the world. One of the important features will undoubtedly be the rapid development of the group for computer education which is affiliated to the society, and has a membership of more than 2,700 secondary school teachers, college lecturers and industrial training officers.

WOLFSON FOUNDATION

Priming the Pump

THE trustees of the Wolfson Foundation are plainly satisfied with the results of the first batch of grants for scientific research linked with industry. A measure of this success is their recent decision to sponsor a further series of projects. The first allocation, in November 1968, (see *Nature*, **220**, 1165; 1968; and **222**, 511; 1969), made available grants totalling £1,055,600 to finance a total of 15 research projects in 10 separate universities. The second allocation, announced in January 1971, provides support for 12 projects in 8 universities at a total cost of £741,730.

The scheme is designed to strengthen links between industry and the universities and was born out of the anxiety which led to the publication of the Docksey report in 1970*. The Wolfson Foundation set up a committee in 1966 to examine the problem and to "give particular attention to such assistance as the Foundation might be able to give to the national effort for the reduction in the time-lag when science is applied to economic purposes". The following year, acting on the advice of the committee, the trustees agreed to make available up to £500,000 a year for five years to support projects, which in the judgment of the trustees might best improve Britain's economic and industrial position. Where this scheme differs from similar projects, like the establishment of the National Research and Development Corporation (NRDC), is the lack of restrictions on the way the Foundation, as a charity, can distribute its money.

The trustees, however, are as financially acute as they are charitable, and they keep a sound weather eye open for a chance of financial reward from their benevolence. The patents rights to any profitable invention, for example, are not waived by the Foundation, and if a university unit started on Foundation capital shows signs of becoming financially viable, then repayment of the grant may be expected, although not demanded; a thoroughly sound arrangement which means that the Foundation can invest this income in other research projects.

*See *Nature*, **227**, 107; 1970.

The first injection of £1,000,000 made possible the start of a wide variety of projects, some of which are now viable self-supporting units. Perhaps the most important—if not the most expensive—schemes involved the establishment of technical service units, units set up in university departments where particular types of information and expertise are available for sale to industry. The best example of this scheme is the University of Southampton where an electronics industrial liaison unit and a centre for industrial noise and vibration control were set up on grants of £24,000 and £30,000 respectively. Both of these units can now balance their books through fees from industry and several other similar units have been established on the campus.

Some projects, of course, although running smoothly have shown no such obvious financial return. Manchester's interest in studies of the stability of earth works by scale models is one example, and the application of modern control theory to models of the national economy at Cambridge is another.

Will the second batch of projects prove as worthwhile as the first? There should be no doubt of this, for the successful twelve were sieved from more than 70 applications by a selection committee headed by Sir Solly Zuckerman. They include a project to develop kenaf as a substitute for jute proposed by Aberdeen and worth £17,500 and the establishment of a centre at Queen's, Belfast to make available to industry the research capabilities of the laser and opto-electronic research group.

Several universities which won grants in 1968 are represented again, including Birmingham and Wales, and the largest single award, £155,850, goes to Newcastle for the establishment of a research and development group for the production and characterization of new metallic and non-metallic materials.

Perhaps the greatest value of the Wolfson scheme, leaving aside the obvious commercial value of the various projects, has been to act as a catalyst. The scheme has awakened wide interest both in industry and in the universities, and now that they have seen how it can be done, many are anxious to attempt similar ventures. The Foundation was, of course, limited in the number of projects it could support by the financial lines drawn by the trustees, but among the 58 proposals rejected this year are many which deserve support, and the Foundation has set the example of how best to apply that support. Perhaps when the trustees come to decide the next grant allocation, they will decide that having provided a working model of academic/industrial cooperation is enough; and they will set their minds to devising other imaginative ways to spend their money.

NEW WORLD

No Cancer Remission for NIH

by our Washington Correspondent

THE manoeuvrings to set up a separate cancer agency outside the National Institutes of Health have entered a delicate stage. Last week the House Appropriations committee approved a proposal to add \$100 million to the budget of the National Cancer Institute, a key stratagem in the Administration's design for buying off the bill being pushed by Senator Edward M. Kennedy, under which a far greater sum would be poured into establishing the National Cancer Institute as an independent agency outside the National Institutes of Health. Another recent Administration stratagem, the promotion of the National Cancer Institute from the status of an institute to a bureau within the NIH, is designed to undermine the Kennedy side's argument that the NIH's cancer effort is superimposed by too many layers of bureaucracy.

This week, just before the meeting of the Senate health subcommittee at which the Kennedy bill will be considered, President Nixon brought the Administration's campaign against the bill to a climax by announcing a new name for the various Administration initiatives, the Cancer-Cure Program. The programme, which is substantially the National Cancer Institute under another name but \$100 million the richer, will remain part of the National Institutes of Health but will be independently budgeted and directly responsible to the President. Nixon also promised that if the extra \$100 million the Administration has requested for cancer this year is not enough, more would be provided next year. These changes are designed to enhance the standing of the National Cancer Institute in every possible way short of removing it from the National Institutes of Health.

The Administration's cause has also been assisted by the response of the biomedical community which in hearings in March before Senator Kennedy's Health subcommittee and subsequently has almost unanimously opposed the plan to take NCI out of the NIH, chiefly on the grounds that other institutes would also want to be independent, which would mean the end of the NIH and the balanced distribution of research funds. In Congress, however, there are few who are willing to cast a vote that might be interpreted as opposed to the conquest of cancer, and the Kennedy bill already has 52 co-sponsors in the Senate. In the House, a lonely but significant voice of

scepticism is that of Representative Paul G. Rogers, chairman of the public health subcommittee which will consider the various cancer bills that have been introduced into the House. Rogers is understood to feel that separating the NCI from the NIH would not necessarily solve any of the present bureaucratic tangles that may exist and could create new problems, such as loss of time while getting the new agency established.

One of the chief arguments of the National Panel of Consultants on the Conquest of Cancer, on whose report the Kennedy bill is based, is that at its present administrative level the National Cancer Institute is buried under multitudinous layers of bureaucracy that stifle initiative from below and dissipate direction from above. Testifying before the Kennedy subcommittee in March, Benno C. Schmidt, chairman of the cancer panel, claimed the institute was buried six levels down in the administrative hierarchy of the Department of Health, Education and Welfare; Carl G. Baker, acting director of the NCI, reports to Robert Berliner, NIH deputy director for Science, who answers to Robert Q. Marston, director of the NIH, who accounts to Surgeon-General Jesse L. Steinfeld, who reports to assistant secretary Roger O. Egeberg, who is answerable to the secretary of HEW, Elliot L. Richardson. Officials at the NIH dispute this chain of command but the Administration has nevertheless reacted to the criticism. Filling in some of the details in the Administration's plan for stonewalling Kennedy, Surgeon-General Jesse L. Steinfeld told the Senate Appropriations subcommittee on Labor and HEW last month that the NCI would be elevated to the status of a bureau within the National Institutes of Health with the director of the new bureau also becoming a deputy director of the NIH. Steinfeld also described the proposed breakdown of the \$100 million supplement requested by President Nixon for the NCI's 1971 budget. Some \$34 million will be assigned to planning, construction and increased operational expenses, \$2 million will be spent on training scientists and technicians, and the rest will go into research.

The \$64 million bonus for research funds, which may be the best thing to come out of the Nixon-Kennedy cancer skirmish, is to be allocated as follows: \$20 million for research in cancer aetiology, of which \$9 million is ear-

marked for viral oncology, \$10 million for chemical carcinogenesis and \$1 million for biometry and epidemiology; \$19 million for research on therapy including radiology and surgery but chiefly chemotherapy—the number of chemical compounds and natural products being screened for anti-tumour activity will be stepped up from 15,000 to 50,000 a year in order to improve on the average of 6 compounds a year currently found worth clinical evaluation; and \$25 million for fundamental research, of which roughly a third will be for individual research grants and two thirds for programme grants.

These funds, which will be spread over a two-year period from July 1, 1971, to June 30, 1973, are in addition to the NCI's regular appropriations, which for the coming financial year amount to \$232 million. The extra \$9 million for viral oncology, a subject whose promise has already been generously recognized in research budgets, probably brings the number of supportable projects in the field close to saturation level. Overall, the extra research funds should allow the National Cancer Institute to fund some 75 per cent of its approved projects—last year the institute was able to finance only 40 per cent of the projects submitted to it and approved as worthy of support.

Of less tangible value—except in so far as it may save the NIH from dismemberment—is the wholly Irish promotion of the National Cancer Institute to the level of a bureau. Asked what difference the new rank would make to its operations, an official of the National Cancer Institute could only think of two administrative privileges involved in the promotion—the NCI will be able to establish the grades at which it hires people, and will be entitled to send out its research contracts without first having them signed by the NIH contract officer. The contract officer is nearly but not quite a rubber stamp—his job is to ascertain that contracts have been properly advertised and evaluated—and skipping him may indeed expedite the processing of NCI contracts by a few days, although NCI officials were unable to estimate what the saving in time will be.

The response from the biomedical community has been fairly united in opposition to the Kennedy plan. The American Cancer Society, three of whose past presidents are members of the cancer panel, naturally supported

the proposal for a separate and more generously endowed agency—the Kennedy bill calls for an initial three-year budget of \$1,200 million. But the only group to support the Cancer Society in testimony before Senator Kennedy's subcommittee in March was the American Heart Association. Campbell Moses, medical director of the Heart Association, told the subcommittee he was in favour of spinning off the NCI as a separate agency and moreover that "an exactly parallel effort is even more appropriate in the field of heart disease". This statement was as neat a demonstration as could be wished of the chief argument adduced by the bill's opponents, that other institutes would want to quit the NIH if the NCI were allowed out.

Critics of the Kennedy proposal included Dr Philip Lee, assistant secretary of HEW for health and scientific affairs during the Johnson Administration, and Dr James A. Shannon, former director of the National Institutes of Health. At the Kennedy subcommittee hearings, Lee produced a letter from Shannon which described the proposed separation of the NCI as "without merit and dangerously destructive". The creation of an independent cancer authority, Shannon said, would represent no improvement on present NIH processes and would "unleash forces of a divisive character which would quickly destroy the integrity of NIH". Shannon went on to predict that "in a very short time, orderly governance would be replaced by anarchy, and instead of a judiciously balanced program of biomedical research, program emphasis would be entirely determined by uncritical zealots, by experts in advertising and public relations and by rapacious 'empire builders'".

Shannon was similarly abrupt in his views of the programmed, NASA-like research methods which the cancer panel has emphasized as a principal selling point of the proposed new agency. Organized research programmes of this type, Shannon said, had been mounted during his time as director of the NIH; they were "encouraged to operate in styles somewhat akin to those employed on the various NASA and big weapons projects", yet despite the managerial competence of those in charge and the size of the investments, "the results have been modest. The reason, as you well recognize, is the inadequacy of the science base."

Other opponents of the Kennedy bill included the Association of American Medical Colleges and the 11,000 member Federation of American Societies for Experimental Biology. John A. D. Cooper, president of the AAMC, told the subcommittee that establishment of a separate cancer authority would duplicate many research teams and

laboratories, which would probably "dilute efforts against other killing and crippling diseases as well as against cancer". And last week a group of nearly 80 professors and department chairmen at medical schools sent a telegram to President Nixon opposing the creation of a separate authority.

The intellectual advantage in the preliminary skirmishing about the proposed new cancer agency probably lies with the Administration forces, but the Administration has now shot most of its bolts and can but wait and see how the proposal fares in Congress. The Kennedy bill is due to be voted on this week by the Senate health subcommittee, several members of whom are said to be opposed to it. If it passes this hurdle, the bill's sponsorship by 52 senators as good as assures its passage through the Senate in some form or other, subject to whatever amendments may be introduced from the floor. Meanwhile the White House, unable to disguise the political rivalry between Nixon and Kennedy which is the principal cause of the conflict, has at least tried to draw a veil over the parallel in-fighting among the scientific

community between cancer researchers and other biomedical scientists. In a letter to the *Washington Post* on March 3, Edward E. David, the President's Science Adviser, said he hoped the public had not acquired any impression that there was an active conflict among scientists over the issue. "There is not now nor will there be, in my opinion, any time or energy wasted on in-fighting. . . ." David assured *Post* readers. "Very few scientists of my acquaintance would participate in such a clearly unproductive activity."

IMMIGRATION

Brain Drain Unplugged

by our Washington Correspondent

RECENT statements that the brain drain of scientists to the United States had halted or was even going into reverse seem to have been premature. The latest figures released by the National Science Foundation show that in the year up to June 30, 1970, the influx of scientists and engineers from other countries was the largest in the past 20 years, and greater by 30 per cent than

New Council for NASA

by our Washington Correspondent

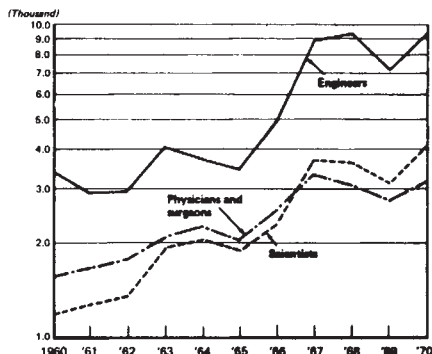
AFTER nearly a year's hiatus, the National Aeronautics and Space Administration has re-established its system of internal science advisory boards. The new arrangement, known as the Space Program Advisory Council or SPAC, consists of a council and four separate committees dealing with applications, physical sciences, space systems, and life sciences (the latter including both space medicine and the search for extraterrestrial life). The council and its four committees replace a considerably more elaborate system of some 15 separate committees such as the lunar and planetary missions board, the astronomy board and other groups of outside scientists who advised NASA on all aspects of the space programme.

The old system was allowed to lapse about a year ago, one of the reasons being a directive from Congress that NASA should cut down on its extensive employment of outside consultants (although scientists were not paid for attending meetings of the advisory boards, travel expenses were considerable). The new system represents an economy, though NASA officials stress that the mixed nature of current and future payloads calls for a wide and interdisciplinary composition of the science advisory boards instead of a batch of specialist committees each dealing with a separate aspect as before.

Members of the new committees, NASA officials say, have been specially chosen for their ability to give advice across a range of fields. Even so, it will be difficult for a single committee to encompass the expertise necessary to advise on the details of individual payloads. In the opinion of one experienced adviser on the space programme, NASA will probably have to recruit further *ad hoc* committees to advise on particular payloads, as was the practice under the former advisory system.

Chairman of the new Space Program Advisory Council is Brian O'Brien, a 73 year old but energetic physics consultant with long experience in advising the Department of Defense on space matters and an expertise in optical physics. Although the new system represents a reduction in the number of scientific advisers NASA can afford, the agency will still have available to it the Space Science Board of the National Academy of Sciences. NASA may well rely increasingly upon the Space Science Board for expert advice, even though the board's most recent report (see *Nature*, **230**, 142; 1971) made major criticisms of priorities assigned by NASA to various aspects of the space programme such as the Grand Tour missions to the outer planets and Planetary Explorer missions to the inner planets.

the influx the previous year. In the fiscal year 1970, 13,337 scientists and engineers and 3,155 doctors entered the United States from abroad. This represents a sharp reversal of the downward trend between 1968 and 1969 when the influx of engineers and scientists dropped from 12,973 to 10,255 and of doctors from 3,060 to 2,756.



Immigrant scientists, engineers, and physicians, by broad occupational group, fiscal year 1960–70.

One reason for the increase is the 1965 revision of immigration quotas whereby quotas from Asian countries were increased at the expense of those from Europe. Engineers and scientists from Asian countries, particularly India, the Philippines, Taiwan and Korea, have pushed up the Asian brain drain from 4,000 in 1968 to 4,900 in 1969 and 7,500 in 1970.

The country most seriously affected by the brain drain is India, which in 1970 lost 2,900 engineers and scientists and 242 doctors to the United States. The second most serious casualty is the Philippines, with a drain of 1,550 engineers and scientists and 770 doctors. In Europe, Britain is still the heaviest loser, being forsaken by 680 engineers, 220 natural scientists, 40 social scientists and 192 doctors, an aggregate slightly greater than that of 1969.

The largest category in the brain drain is that of the engineers, who last year formed 9,300 or about 70 per cent of the total. Within this category, fewer aeronautical engineers entered the United States last year than in 1969, but all other engineering specialties flocked in in greater numbers, making an overall increase of 30 per cent. The influx of natural scientists swelled by 25 per cent to 3,260, and at 770 the number of social scientists was half as large again as the inflow in 1969.

Since February this year immigrants to the United States have been required to have a job offer for which Americans are not readily available. This condition may not reduce the brain drain in any serious way but will merely tailor the rest of the world's loss of scientific manpower more closely to the needs of the United States job market.

Short Notes

by our Washington Correspondent

Sickle Cell Anaemia

LEGISLATION to set up a new institute of the National Institutes of Health devoted to sickle cell anaemia was proposed recently by Representative Ella T. Grasso. The disease, which affects one in every 400 to 500 blacks, kills half its victims before the age of 20. The theoretical basis of the disease is rather well understood. Ingram's discovery of the single amino-acid change in sickle cell haemoglobin was one of the earliest triumphs of molecular genetics—but medical interest in developing a practical treatment has been curiously slow.

The NIH's most recent contribution to the field was a research grant in support of a procedure, widely publicized as the first "fully effective" treatment, which consisted of injecting high concentrations of urea intravenously. The budget statements prepared by the Nixon Administration last February promised that research on sickle cell anaemia would be "substantially expanded". President Nixon was even to have mentioned the disease in his State of the Union message but the reference was dropped from the final draft, apparently on the grounds that sickle cell anaemia is not well known among the general public.

More recently the administration announced a pilot genetic counselling programme to advise prospective marriage partners among the 10 per cent of the black population that carries the sickle cell gene of the genetic risks involved.

Lost Nerve Gas

THE US Army is still in trouble over the matter of 200 canisters of VX nerve gas it managed to lose in a small Alaskan lake in 1966. The canisters, scheduled for destruction, were stored on the frozen surface of a lake in the army's Gerstle River test area, but the order for destruction never came. One sunny day in May, the lake thawed and the canisters sank to the bottom, leaving only confused rumours of their existence. No records of the canisters' fate being maintained, it was not until two years later that the army thought of draining the lake to see if the rumours were true. By August 1968 the recovered munitions were being disarmed, and nine months later the army got around to announcing the incident. Local residents had not been warned at the time, it was explained, because the lake site was remote from populated areas and there was no sign that any of the canisters had leaked. The incident might have been forgotten as just another example of chemical and

biological weapons being handled in cavalier fashion, but that two soldiers stationed at Fort Greeley, the Alaskan test area, died during the time the nerve gas canisters were being recovered from the lake. Pneumonia, the army said. Recently, Senator Mike Gravel of Alaska wrote asking for a fuller account to the Secretary of Defense. The doctor attending one of the soldiers, Gravel's letter stated, had written in a progress report that he didn't think the illness was due to pneumonia, and one of the soldier's assignments had been cleaning gas-masks.

Pot Will Follow Porn

THE high value attached by President Nixon to rational inquiry as a means of arriving at solutions was amply demonstrated by his reaction to the report of the Commission on Obscenity and Pornography which reported last year. Further demonstration was provided two weeks ago at a news conference held at San Clemente, California. Asked about the question of legalizing marihuana, a subject now under study by a presidential commission, Nixon stated, "I am against legalizing marihuana. Even if the commission does recommend that it be legalized, I will not follow that recommendation." Like the judge in the Manson case and the review board in the Calley case, the commissioners investigating marihuana now know to what conclusion the evidence must lead them.

Chemists Out of Work

SOME 5,000 of the nation's 186,000 chemists and chemical engineers are now out of work, according to a survey conducted by the American Chemical Society, and another 12,000 are in varying states of insecurity ranging from temporarily unemployed to part-time employment and post-doctoral positions. The ACS survey estimates that at least 11,000 of these 17,000 need new jobs but they will be competing with the 1971 crop of graduates—16,000 bachelors, 3,000 masters and 3,000 PhDs, at least half of whom, or about 11,000, will be actively seeking immediate professional employment. The number of jobs needed for chemical scientists in the coming months could thus rise to some 22,000, which is more than double the number of jobs available in a normal year.

The estimates of unemployment are based on a survey taken among ACS members; 2.7 per cent of the 27,000 members responding reported they were unemployed as at March 1, 1971. Unemployment was worse than average among those under the age of 25, seven per cent of whom were jobless, and among women chemists, of whom 6.3 per cent were unemployed.

NEWS AND VIEWS

Revolutionary Activity in the Upper Atmosphere

HUMAN beings have grown used to breathing, and do not often remind themselves that if the Earth were less massive, all its vital air would escape into space. There is just as much complacency about atmospheric rotation. It is blithely assumed that the air-blanket will go on spinning at nearly the same rate as the Earth, with a velocity approaching 500 m/s at the equator; it is difficult to credit the idea that the atmosphere might flip over into a regime with an unearthly rotation rate, and subject everybody to a perpetual hurricane.

Consequently most people have a built-in prejudice that the upper atmosphere co-rotates with the Earth, and there was scepticism when measurements of the orbits of the first satellites indicated that the upper atmosphere at heights near 250 km might be rotating faster than the Earth (*Proc. Roy. Soc., A*, **253**, 529; 1959). In the past five years the average rotational speed of the upper atmosphere has been determined from the changes in the orbital inclinations of many satellites, and the most recent review by King-Hele, Scott and Walker (*Planet. Space Sci.*, **18**, 1433; 1970) indicates that the average angular velocity of the upper atmosphere increases from 1.1 times the Earth's angular velocity at a height of 200 km to 1.4 times at a height of 350 km. At latitude 30° this corresponds to mean west-to-east wind speeds of 40 m/s at 200 km height, increasing to 160 m/s at 350 km. These studies should soon be improved in accuracy and extended to greater heights now that numerous observations are becoming available from the highly accurate Hewitt cameras at Malvern and Edinburgh.

Upper atmosphere winds are likely to vary greatly between day and night, and with solar activity. Thus if the average zonal wind speed is of the order of 100 m/s, the maximum winds are probably much stronger and have a profound influence on the physics of the ionosphere. Consequently there have been numerous attempts to solve the rather intractable equations of motion. The chief driving force for the winds is usually taken to be the pressure gradient between the daytime maximum pressure, near the equator at about 14 h local time, and the lower pressure in regions where the solar elevation angle is lower. This wind-generating force is opposed by ion drag, the tendency of the ions to collide with and damp down the flow of the neutral particles. Since the ions are generally more numerous by day than at night, most theoretical studies give stronger winds by night than by day. Otherwise there is not much agreement between the studies. For example, Kohl and King (*J. Atmos. Terr. Phys.*, **29**, 1045; 1967) deduced winds of order 100 m/s but very little "super-rotation", whereas studies by Challinor (*Planet. Space Sci.*, **17**, 1097; 1969; **18**, 1485; 1970) gave a substantial excess rotation in equatorial regions. Recently Rishbeth (*Nature*, **229**, 333; 1971; also *Planet. Space Sci.*, **19**, 357; 1971) has emphasized the probable contribution of equatorial F-layer polarization.

Atmospheres that rotate with indecent rapidity also arouse interest among non-atmospheric physicists, as seen in the article by Weinstein and Keeney on page 109 of this issue of *Nature*. They point out that a small cylinder

suspended inside a larger cylinder filled with a rarefied gas suffers a rotational torque in the presence of a radial temperature gradient and an axial magnetic field, though it is doubtful whether this simple phenomenon is relevant to the much more complex conditions prevailing on the Earth.

Nevertheless, the phenomenon of super-rotation may not depend on the particular conditions on Earth. The upper atmosphere of Venus seems to rotate once every 4 or 5 days, equivalent to a mean wind speed of about 100 m/s at the equator. This is super-rotation *par excellence* because the planet itself takes 244 days to rotate. Halley once suggested (*Phil. Trans. Roy. Soc.*, **16**, 153; 1686) that terrestrial trade winds might be caused by rectified motion generated by the diurnal solar heating. This old idea has been pursued and applied to Venus by Schubert and Whitehead (*Science*, **163**, 71; 1969), who found that when a flame was moved round under an annular pan of mercury, the rectified flow could be several times faster than the speed of the heat source. Quite a few theoretical explanations of the so called Halley effect have now been propounded, including those by Schubert and Young, by Malkus and by Thompson (*J. Atmos. Sci.*, **27**, 523, 529 and 1107; 1970).

Jupiter may also exhibit super-rotation, because its atmosphere rotates more rapidly in a narrow equatorial band than at other latitudes (see *J. Atmos. Sci.*, **26**, 841; 1969). A similar situation may prevail on Saturn, and even the quicker rotation of the Sun near its equator may possibly be relevant.

The reader of the many theoretical papers on atmospheric rotation may complain that more facts and less speculation are needed. In the Earth's upper atmosphere this demand is not easily satisfied. The observational results from satellite orbits have so far been poor in time resolution. The other principal observational method is to release reactive substances from high-altitude rockets and measure the motion of the glowing cloud of contaminants. This method suffers from just the opposite defect: it gives only local wind speeds, and only for a few minutes. The favourite material for such experiments used to be sodium, which glows only when sunlit, thus further limiting the observations to morning and evening twilight. In the past few years this limitation has been overcome by using trimethyl aluminium, which glows in the dark; and the technique is also being extended for daylight use. The results from vapour trails at heights above 180 km, though limited in number because of the expense of such launchings, do indicate west-to-east wind speeds of order 100 m/s in evening twilight (for example, *J. Geophys. Res.*, **75**, 683; 1970), but with rather varied winds, mainly east-to-west, in morning twilight. The wind profiles seem to depend not only on time of day, but also on latitude, geomagnetic conditions, and probably many other parameters. For example, some notable results have recently been given by Rees (*J. Brit. Interplanet. Soc.*, **24**, 233; 1971) from firings at the Kiruna range in Sweden, in the auroral zone. Rees found that the west-to-east neutral wind at 150 km height was proportional to the degree of geo-

magnetic disturbance, and winds up to 500 m/s were measured at a latitude where the Earth's rotational speed is only 170 m/s. Such winds are presumably caused by acceleration of the neutral air by the auroral electrojet, a mechanism which could contribute to the super-rotation, as suggested by Cole (*Planet. Space. Sci.*, **19**, 59; 1971).

If progress is proportional to the number of theoretical

studies, the revolutionary behaviour of the upper atmosphere should soon be understood. Detailed observational results from vapour trails may continue to be rather sparse, not only because of the expense, but also possibly because people may begin to wonder whether the pollution of the upper atmosphere by metal atoms is as harmless as the experts say.

Reverse Transcriptase in Human Milk Virus

At the end of February, Moore and his collaborators in Camden, New Jersey, Detroit and Bombay electrified the worlds of tumour virology and cancer research with their report of the detection in human milks of virus particles virtually identical in structure to mouse mammary tumour viruses (see *Nature*, **229**, 593; 1971). The occurrence of these particles in the milks of a small sample of American women was found to be highly correlated with familial histories of breast cancer. And so it now seems highly likely—although it will be very difficult to prove rigorously—that human breast cancer is at bottom a virus disease, the course of which in any particular woman is strongly influenced by her sex hormone status. This belief receives support from the experiments of Schlom, Spiegelman and Moore reported on page 97 of this issue of *Nature*.

Collaborating with the Camden, Detroit and Bombay teams, Schlom and colleagues have quickly done what was perhaps the most obvious next step towards characterizing the putative human mammary tumour virus; they have assayed milks for RNA dependent DNA polymerase activity, reverse transcriptase, and correlated the presence or absence of this enzyme with the presence or absence of the virus particles. They have come up with the predictable result; of the thirteen milks they tested, only four were found to have this activity and only these four milks, as the Camden group found, contain detectable amounts of human mammary tumour virus. Furthermore, the reverse transcriptase activity is uniquely associated with particles which sediment on centrifugation in density gradients at a density of 1.17 to 1.19 g per ml., the density of the virus particles. In other words, the correlation between the presence of the virus and the presence of this enzymatic activity, which has now been found in every sort of tumour

virus which has been assayed, twenty-seven types in all, is perfect in this small sample.

No doubt because this is precisely the result anticipated by everybody, Schlom *et al.* took great pains to obtain unambiguous data. They have, for example, used the original assay system for reverse transcriptase devised by Temin and Mizutani, which involves treating the samples with a non-ionic detergent to disrupt intact virus and then allowing the reverse transcriptase to use the endogenous RNA as template. In these conditions DNA synthesis depends on a supply of the four deoxyribonucleotide triphosphates and it is inhibited by ribonuclease. As Schlom and colleagues point out, they refrained from using synthetic double-stranded RNAs or synthetic RNA/DNA hybrids as template, for, even though these molecules are more efficient templates than viral RNA, they are less specific. Enzymes other than reverse transcriptase can use them to make DNA. The possibility that the activity detected in the four milks arises from contaminating mycoplasma has also been carefully ruled out. Samples of pure human and animal mycoplasmas do not contain reverse transcriptase detectable in the assay used.

Clearly the presence of reverse transcriptase in these human milk virus particles does not prove that they are oncogenic; that will depend on collecting a variety of circumstantial evidence. But the fact remains that these particles are under the gravest suspicion and it is up to the tumour virologists to assemble a totally incriminating case. If they can prove that the human milk viruses are serologically related to murine mammary tumour viruses, that they induce mammary tumours in animals and transform at least some cells *in vitro*, and that the correlation between the presence of the virus in milk and the

risk to breast cancer holds true of large samples of women, few will be left doubting the assertion that breast cancer is a virus disease.

GEOMAGNETIC REVERSALS

Matuyam Reassessed

from our Geomagnetism Correspondent

IT has been evident for some time that although the principal features of the geomagnetic polarity-time scale are well established the finer details are likely to prove troublesome. Part of the reason for this is, of course, that potassium-argon dating—like any other experimental technique—has a limited resolution, which means that the shorter magnetic “events” within the longer and well defined “epochs” are difficult to define precisely. This point is particularly relevant in the light of Cox's theoretical prediction (*J. Geophys. Res.*, **73**, 3247; 1968) that during the past 10 million years, at least, there have been numerous short events with durations less than 0.05 million years. But even longer events are not easy to tie down.

One particular part of the polarity-time scale covering the past four million years or so which has recently proved to be a source of controversy is that in the vicinity of the Olduvai and Gilså polarity events within the Matuyama reversed epoch—that is, the period between approximately 1.6 and 2.1 million years ago. The confusion inherent in this part of the scale is well illustrated by reference to a single author. Three years ago, in a version of the reversal-time scale derived only from dated continental rocks, Cox *et al.* showed (*Quart. J. Geol. Soc. Lond.*, **124**, 53; 1968) the Gilså event lasting from 1.6 to 1.65 million years and the Olduvai event from 1.88 to 2.0 million years. In a more recent version, however, modified in the light of data from deep sea sediment cores and ocean magnetic profiles Cox showed (*Science*, **163**, 237; 1969) each event to be split—1.61 to 1.63 and 1.64 to 1.79 million years for the Gilså and 1.95 to 1.98 and 2.11 to 2.13 for the Olduvai. Moreover, in the second article Cox identified the large event in this region, the only event

normally resolved in deep sea cores and ocean magnetic profiles, as the Gilså at about 1.7 million years rather than, as before, the Olduvai at about 1.95 million years. Emilia and Heinrichs have also suggested (*Science*, **166**, 1267; 1969) that on the basis of uniformity in sea floor spreading rates the long event is the Gilså.

So what is the truth of the matter? Or rather, what is the best interpretation to be derived from currently available data? To try to settle this question Grommé and Hay have returned to look more closely at the Olduvai type section at Olduvai Gorge, Tanzania (*Earth Planet. Sci. Lett.*, **10**, 179; 1971). They show that the normal geomagnetic polarity event is represented there by five normally magnetized volcanic units—two lava flows and three ash flow tuffs. Because of the particular problems involved in the dating of these samples the time range they represent may be as little as 1.70 to 1.75 million years or as great as 1.6 to 1.9 million years. But what is certain is that a reversely magnetized tuff immediately below the normal section sets an earlier limit of 2.0 million years for the beginning of the Olduvai event. In short, on this basis the Olduvai event is no older than 2.0 million years and covers a period of no more than 0.3 million years. If the Olduvai event is limited to 1.6 to 1.8 million years, however, it is possible to assume greater linearity of both deep sea sedimentation rates and sea floor spreading rates.

Grommé and Hay thus suggest that the normally magnetized Icelandic flow dated at 1.65 ± 0.05 million years, which was first used to detect the Gilså event, could well have been produced during the Olduvai. In this case, of course, the Gilså becomes a non-event. On the other hand, there is a little evidence from a few deep sea cores that a very short event occurs just after the longer (Olduvai) one; and so it seems reasonable under the circumstances to regard this as the true Gilså.

But are there any other normal events within the Matuyama epoch? In their 1968 paper, Cox *et al.* listed seven normally magnetized lavas in the range 1.95 to 2.09 million years. In the light of their previous conclusions Grommé and Hay now suggest that some or all of these flows were probably erupted before the Olduvai as they define it. Moreover, there is some evidence from ocean magnetic profiles of one, or possibly two, short normal events preceding the Olduvai. Although it seems probable that there are, in fact, two events here they cannot be distinguished unambiguously from each other by radiometric dating alone; and so Grommé and Hay propose that they be called the Réunion events because they are best represented on Réunion Island.

MARINE BIOLOGY

Death in Massachusetts

THE news of mass mortalities of marine animals tends nowadays to be ammunition for the anti-pollution lobbies. But all too easily it is forgotten that such occurrences often happen quite naturally as a consequence of extremes of temperature, low salinity and the like. They often pass unrecorded, but one instance of a natural mass mortality involving a type of fish known as the crevalle jack (*Caranx hippos*) caught the attention in 1969 of J. G. Hoff of Southeastern Massachusetts University.

In a brief note in *Chesapeake Science* (**12**, 49; 1971) he reports seeing on October 18, 1969, more than two hundred dead crevalle jacks in the upper reaches of the Slocum River in Massachusetts. He collected at random some fifty-five dead fish, with an average length of 3.8 inches, but he regards his estimate of the number of dead as being on the low side because he spotted about twenty herring gulls feeding on the dead fish.

Juvenile crevalle jacks are annual

visitors to the Slocum River—they usually appear in the river in mid-June and leave by mid-October for southern seas. In more than two years of surveys in the Slocum River, Hoff had recorded neither pollution nor mass mortalities of fish. But where he found the dead crevalle jacks in 1969, the temperature measured only $7.4\text{--}9.0^\circ\text{C}$; farther downstream where the temperature ranged from $9.8\text{--}10.9^\circ\text{C}$, the fish were living quite normally. Salinity was low in the cold area, but this was not unusual and crevalle jacks had survived it in the past.

The crevalle jack seems to have been the only species of fish affected by the low temperature in the area, for several other species were caught alive with a haul seine. Hoff concludes that the cold microclimate in the Slocum River in October 1969, caused by the influx of cold spring water from a tributary combined with a sudden drop in air temperatures, killed the crevalle jacks and he says that it is likely that other late crevalle jack migrants meet the same fate in cold northern waters in the open sea.

Infrared Background points to Low Density Universe

WITH the discovery of a strong background of infrared radiation, it seems that most arguments against a universal origin for cosmic rays can be countered, provided that the density of gas and dust between the galaxies is very low. In *Nature Physical Science* (Monday, May 17) G. Setti and L. Woltjer develop the argument first put forward a few years ago by Setti and M. J. Rees, that the energy density of the universal background of X-radiation is linked to that of the extragalactic cosmic rays through the inverse Compton effect. When this idea was first suggested (*Nature*, **219**, 127; 1968), it seemed that the sources of energy available to drive this process—strong extragalactic radio sources—were not sufficient to account for the observed flux of cosmic rays. But now the discovery of powerful infrared sources has provided an additional energy source, which may tip the balance in favour of the inverse Compton process. This mechanism, by which energetic electromagnetic radiation interacts with charged particles to produce fast particles and weak radiation, is thought to operate in many astronomical objects.

The mechanism driving the infrared sources is not known, but it is sufficient for the simple model of Setti and Woltjer that the energy is being produced, and that there must certainly be electrons, protons, and magnetic fields somewhere around the sources, so that the acceleration of particles can occur. Perhaps the most interesting aspect of this latest work is the conclusion which

is drawn concerning the density of matter between the galaxies. Certainly a universal cosmic ray flux will be affected by its passage through this matter, losing its energy by the production of energetic photons. The most generous estimates suggest that only a few times 10^{-7} particles per cubic centimetre would be sufficient to absorb almost all the cosmic rays; even the heating of such a tenuous gas to 10^9 K by the cosmic ray particles would not produce detectable re-emission of the energy. The resulting ionization of the gas would, however, explain why no Lyman alpha absorption is seen in the spectra of quasars.

So although the arguments are still finely balanced, it does seem that the energy requirements for a universal origin of cosmic rays can be met. The one factor which seems implausible on some theoretical grounds—the required low density of intergalactic matter—neatly explains two observational problems: the lack of Lyman alpha in quasar spectra and the lack of any other direct detection of matter between the galaxies. But most intriguing of all, if there is so little matter between the galaxies the mechanism by which galaxies form must therefore be very efficient. It is hard to see how so little matter could be left over from any conventional collapsing gas cloud model for the origin of galaxies, and this is bound to raise once again the question of galaxy formation from the inside outwards (by the expansion of a “white hole”).

SUPPRESSION

***Drosophila* Mutations**

from our Cell Biology Correspondent

BIOLOGISTS who enjoy vicarious pleasures should not miss reading the current issue of the *Journal of Molecular Biology* (57, 231; 1971), for in it Twardzik, Grell and Jacobson report that a suppressor mutation, which suppresses the vermilion eye colour mutation of *Drosophila melanogaster*, alters a species of the fly's tyrosine tRNAs.

Mention of genetic suppression to anybody reared on the biology of *Escherichia coli* conjures up, of course, pictures of mutated transfer RNA molecules which, either because they have a new anticodon, or as a result of some other structural modification, have gained the ability to read nonsense chain terminating codons or to rectify missense mutations. Fully aware that suppression in microorganisms is usually mediated by a structurally changed tRNA, Twardzik and his colleagues simply asked if the same might not be true of suppression in higher organisms, about the mechanism of which nothing is known; their hunch has paid off handsomely, albeit in an unexpected way.

Geneticists long since isolated a recessive sex-linked suppressor mutation, *su(s)^h*, which is non-allelic to the sex-linked vermilion eye colour mutation of *Drosophila*, which it suppresses. *Drosophila* carrying the vermilion mutation lack a brown eye pigment because they are unable to convert tryptophan to kynurenine, an intermediate step, catalysed by tryptophan pyrrolase, in the synthesis of the brown pigment. In these mutant flies tryptophan pyrrolase activity is never more than 25 per cent that in wild type flies but when the *su(s)^h* suppressor mutation is introduced the activity of this enzyme is partially restored, the large accumulation of non-protein tryptophan, which characterizes the vermilion mutants, is reduced, and eye colour is restored.

Taking the bull by the horns, Twardzik *et al.* screened extracts of flies, searching by reverse phase chromatography for a modified species of tRNA in the *su(s)^h* homozygotes; and sure enough they found that the *su(s)^h* locus controls directly the amount of a species of tyrosine tRNA in adult flies. Wild type *Drosophila* have three resolvable tyrosine tRNAs, two major species and a minor, and *su(s)^h* flies lack the second major species and have a larger than normal amount of tRNA chromatographing at the position of the first major species. Flies bearing a new suppressor mutation *su(s)^{e1}* isolated by Twardzik *et al.*, which maps at the same locus as *su(s)^h*, also lack the second major tyrosine tRNA. A series of genetic crosses established that although

the vermilion and suppressor loci are both on the X chromosome, only the suppressor locus controls the loss of the tyrosine tRNA. The recessive nature of the suppressor mutations and the fact that tRNA structural genes are redundant in eukaryotes indicate that the suppressor locus is not the structural gene for the tRNA in question but the suppressor locus presumably specifies an enzyme which somehow modifies this tRNA during its maturation.

But how does the loss of the second tyrosine tRNA, tRNA^{Tyr}, suppress the vermilion mutation? At the time Twardzik and his colleagues wrote their report they were clearly thinking along conventional lines, speculating about possible ambiguous codon responses of the unmodified tRNA^{Tyr}, and suppression at the level of translation. But further experiments reported recently by Jacobson in *Nature New Biology* (231, 17; 1971) have changed all that. Jacobson has found that by treating homogenates of vermilion mutant flies with ribonuclease T1 he was able to restore their tryptophan pyrrolase activity. The obvious implication was that an RNA inhibits tryptophan pyrrolase in vermilion mutants. Pursuing this lead

he has shown that the addition of uncharged tRNA^{Tyr} to the tryptophan pyrrolase of vermilion mutants, activated by ribonuclease digestion, once more inhibits the enzyme's activity. This species of tRNA is therefore an inhibitor of the vermilion mutant enzyme but it does not inhibit wild type enzyme.

From all this Jacobson argues that the wild type enzyme is associated in some way with tRNA^{Tyr} and is active whereas the tryptophan pyrrolase specified by the vermilion locus, presumably because of its mutated structure, is inhibited when it associates with this tRNA. Removal of the tRNA either by nuclease digestion or by introducing the suppressor *su(s)^h* mutation, which prevents the maturation of tRNA^{Tyr}, leads to the reactivation of the mutant enzyme. Clearly this is the first of a fascinating collection of stories. Why so many isoaccepting species of tRNAs exist has long been a puzzle but there is now a clue as to their function. They may well be more important for the control of enzyme activities—in other words the control of metabolism and differentiation—than the control of translation.

Intricacies of Starting a Protein

In *Nature New Biology* next Wednesday, Rudland, Whybrow and Clark suggest that a protein called initiation factor F2 plays a crucial part in ensuring that the transfer RNA carrying the amino-acid which initiates the synthesis of all *Escherichia coli* proteins goes to the correct site in a ribosome.

During protein synthesis each transfer RNA molecule picks up a particular amino-acid, carries it to the ribosome and, if the codon for that amino-acid is waiting to be read, delivers it to the site at which amino-acids are added to a growing polypeptide chain. Clearly, ensuring that the charged tRNA is delivered to precisely the right site in the ribosome is of crucial importance and it is known that a protein called T factor promotes this reaction.

T factor will bind with a molecule of charged tRNA and a molecule of GTP to form a complex; this complex then delivers the tRNA complete with its specific amino-acid to what is called the A-site of a ribosome. The A-site is the reception centre for charged tRNA molecules which can donate the amino-acid they carry to the growing protein held in the so-called P-site.

But T factor only promotes the delivery of charged tRNAs to a ribosome which has already started to synthesize a protein. According to

Rudland and his colleagues a different factor, the F2 initiation factor, is involved in the delivery of the very first charged tRNA which initiates protein synthesis. They have shown that F2 protein will form a complex with a molecule of GTP and a molecule of the initiator transfer RNA charged with the initiating amino-acid, formylmethionyl-tRNA_f. They envisage that F2, in this complex, plays a part analogous to T factor, but instead of delivering the formylmethionyl-tRNA_f to the A-site of a ribosome it delivers the initiator to the P-site.

Once the formylmethionyl-tRNA_f is bound to the P-site the second and subsequent amino-acids attached to their tRNAs can be delivered by the T factor to the A-site and added to the growing chain. And to ensure that only the initiator species of transfer RNA enters the P-site the two factors T and F2 have evolved mutually exclusive specificities. T factor will complex with any charged tRNA except the initiator species, while factor F2 will only complex with, and therefore deliver, the initiator formylmethionyl-tRNA_f. Having hit on a successful basic mechanism for the delivery of charged tRNAs to ribosomes, nature has evolved two neat variations to make certain that the processes of starting a protein and elongating a protein do not interfere with each other.

CONTINENTAL DRIFT

Dissent on Split

from our Geomagnetism Correspondent

IN a sharply worded and provocative note (*Earth Planet. Sci. Lett.*, **10**, 271; 1971) Wright takes issue with Larson and LaFontain who recently suggested (*Earth Planet. Sci. Lett.*, **8**, 341; 1971) on the basis of palaeomagnetic evidence that both the North Atlantic and South Atlantic began to open up about 200 million years ago. The burden of Wright's criticism is aimed directly at Larson and LaFontain; but rightly or wrongly he chooses to extend his target to geophysicists in general and what he clearly takes to be their arrogance in apparently refusing to take full account of geological data when it comes to such things as continental drift. Whether the imputation of guilt by association will be taken humbly by geophysicists remains to be seen.

In their original article Larson and LaFontain made an attempt to test the continental fit of Bullard *et al.* (*Phil. Trans. Roy. Soc., A*, **258**, 41; 1965) using up to date palaeomagnetic data. Their technique was to hold North America stationary and rotate the other continents into the relative positions indicated by the Bullard reconstruction. When this was done, Carboniferous poles from Eurasia and North America, which were widely separated before the rotation, moved to near coincidence. So did Permian poles from North America, Eurasia and Africa. From this Larson and LaFontain inferred that during the Carboniferous and Permian, Europe, North America and Africa were contiguous, or almost so. The situation with regard to South America was less certain; but in so far as the few available poles moved towards the common pole when rotated, the same conclusion could, according to the authors, justifiably be drawn.

In the Triassic the polar coincidences after rotation were not quite so good, which seemed to indicate that this was when the continents began to split. This was confirmed by the behaviour of the Cretaceous poles. Rotation of the African poles to the Bullard fit produced first a convergence and then an increased scatter—an effect to be expected if the rocks had been magnetized since drift began. Moreover, the closest coincidence of North American and African Cretaceous poles occurred about halfway along the rotation path which, according to Larson and LaFontain, must mean that by the Cretaceous the South Atlantic had already opened up by about 50 per cent. Rotation of the Eurasian poles, on the other hand, produced divergence right from the start. Thus by about 100 million years ago the North Atlantic above 40° N

must have been about as wide as it is now.

Wright's general objection to all this is "that in 1970 it is still possible for geophysicists to make pronouncements about Earth history without considering geological evidence at all". More particularly he points to evidence already published which suggests that the South Atlantic, at least, must have opened much later than the Triassic. The distribution of salt deposits on both sides of the South Atlantic, for example, has apparently shown that marine conditions did not reach positions about halfway up Africa and South America until about 110 million years ago. Palaeontological evidence from Brazil and West Africa also suggests a final separation of the two continents about 90 million years ago. And the tectonic history of the Benue trough in Nigeria, in turn, gives a split about 80 million years ago.

In quoting this specific evidence Wright is on pretty strong, if somewhat qualitative, ground and would probably

have been wise to leave it there. His more general pronouncements are, however, altogether more contentious. He believes, for example, that "geological criteria are *still* (Wright's italics) the most reliable when it comes to reconstructing pre-drift configurations and establishing when they broke up". The irony of this statement becomes apparent when it is remembered that geological evidence was available for at least the first half of this century and yet failed to convince all but a handful of geologists that continental drift was anything but science fiction. Then again he quotes a statement from a colleague in the United States to the effect that "95% of geologists would agree that the South Atlantic opened around 110 million years ago and not 200 million years". To which the only adequate reply would seem to be that thirty years ago 95 per cent of geologists would have agreed that the South Atlantic never opened at all—it was there since creation.

Carbonic Anhydrase may Regulate Photosynthesis

Now that the dust has, to a large extent, settled on the battles fought over the mechanisms of photosynthesis, the centre of interest seems to have shifted from carbon pathways and electron movements to the problems of how the process is regulated. In next week's *Nature New Biology*, D. Graham and M. L. Reed suggest that carbonic anhydrase, an enzyme which facilitates the combination of water and carbon dioxide to yield HCO_3^- and free protons, may play an important, if not the most important, part in the control of photosynthetic processes.

On the face of things this may seem an ambitious proposal, but few workers will deny that enzymologists have always had the happy knack of spinning the most remarkable webs of biochemical pathways on the evidence of finding unexpected enzymatic activities in plant or animal systems. The new hypothesis is based on an unusual photosynthetic phenomenon which can be observed in the unicellular green alga *Chlorella pyrenoidosa*. Cells grown in high partial pressures of CO_2 (5 per cent by volume) show very low rates of photosynthesis if transferred to a CO_2 concentration some ten times less than that in air. An induction period of about 2 hours seems to be necessary before the normal Calvin cycle of CO_2 fixation is restored. In contrast, cells grown in air (0.03 per cent CO_2) do not show this lag on transfer to a low CO_2 environment.

Graham and Reed discovered that the level of carbonic anhydrase activity was very low in cells grown in high CO_2 , but that the activity of this enzyme

slowly increased, concomitantly with photosynthetic activity, when the cells were transferred to low CO_2 . Furthermore, this adaptation to the low CO_2 environment could be inhibited by a prior treatment with 'Diamox', a specific inhibitor of carbonic anhydrase. On the other hand, cells grown in high CO_2 showed no depression in photosynthetic activity if this was measured in high CO_2 , even if 'Diamox' was present.

What all this suggests is that the rate of photosynthesis is regulated by the activity of carbonic anhydrase when the concentration of CO_2 is limiting. How this regulation may be achieved is not yet certain, but Graham and Reed have a novel proposal. They suggest that the enzyme may make available the protons which are required to maintain a pH gradient across the thylakoid membranes of the chloroplast, or to regulate the proton gradient in the chloroplast. Such a pH gradient is, of course, an essential feature of photophosphorylation according to Mitchell's chemiosmotic hypothesis.

Of course, other functions for carbonic anhydrase can be put forward—Graham and Reed mention the possibilities of increasing the concentration of free CO_2 at the site of carbon fixation or changing the permeability of the thylakoid chloroplast membranes—but evidence for these ideas is as scarce as it is for Graham and Reed's hypothesis. And of course the Mitchell hypothesis has not yet found universal acceptance—persuasive as the carbonic anhydrase scheme appears, it is unlikely to prove the last word in the regulation of photosynthesis.

RNA STRUCTURE

Loops and Pairs

from our Molecular Biology Correspondent

WHEN a few years ago the sequences of low-molecular weight RNAs began to appear, they were seized on by eager individuals, who, by pure contemplation, by model building or by computer methods, set off in pursuit of convincing base-pairing schemes. Before long a horrible murrain of hypothetical structures had spread through the literature. So far as any lessons were to be drawn from them, it was that in general—and the cloverleaf model by tRNA is probably an exception—there are as many feasible base-pairing schemes as there are authors. If then one's interest in predicting the base-pairing schemes from sequences is anything more than frivolous, there is little choice but to lay the ground rules by the use of specially tailored models. This of course is hard work, but as two new articles from Doty's laboratory at Harvard demonstrate, the effort is amply justified.

The difficulty that has beset the study of base-pairing in oligomers of A and of U, for example, has been the preferential formation of three-stranded structures. To overcome this problem Martin, Uhlenbeck and Doty (*J. Mol. Biol.*, **55**, 201; 1971) have synthesized block oligomers, in which a short run of U residues is grafted on to a similar run of A. The formation of three-stranded helices, (A+2U), from these species is scarcely feasible if the lengths of the blocks are similar, and indeed the spectroscopic evidence is strong in favour of double strands. Moreover, the melting curves in these systems show a strong concentration dependence, which indicates that association occurs between two molecules, and not by the formation of hairpins, which would preclude the pairing of as many residues as are required to make the turn. From the concentration-dependence of the melting temperature, a theoretical treatment worked out by Applequist can be used to derive the thermodynamic parameters associated with base-pairing. These include the enthalpy per base-pair, which turns out to depend on the helix length—a result which is not altogether unexpected, and may arise from a lesser contribution from the bases at the ends of the short duplex, or from the increased magnitude of the stacking interactions in the melted state, when the melting occurs at lower temperature.

When a solitary C is introduced between the A and U blocks, the double helix is interrupted in the middle by an unpaired residue in each strand, and there is accordingly a sharp decrease in stability. This type of effect is further pursued in a second paper (Uhlenbeck, Martin and Doty, *ibid.*, 217). In the

first place they show that the effect of an unpaired G is much less severe than that of a C. This at last lends concrete substance to the widespread belief in the existence of G-U base-pairs: two antiparallel strands of the oligomer A_nGU_n can form two G-U pairs and six A-U pairs with an overlap of one unpaired A at either end—a scheme which is supported by the similar stability of chains in which the G is off-centre. The argument is that if the G residues were looped out of the helix, the effect of two single-base loops would assuredly be different from that of one bulge in the centre with two G residues opposed. When, on the other hand, a G-C pair is inserted, as in complexes of A_nGU_n with U_nCA_n , there is a large increase in stability, and this increases further in self-paired A_nGCU_n . A G-C pair is evidently worth 3-4 kcal/mole more than an A-U, and it is composition more than length which dominates the helix stability. A graphic illustration of the latter principle is also given

by Coutts (*Biochim. Biophys. Acta*, **232**, 94; 1971), who finds a very high melting temperature for a fragment of tRNA containing four G-C pairs only.

To appreciate the value of data of this kind, one need only now examine the paper by Tinoco, Uhlenbeck and Levine in *Nature* last month (**230**, 362; 1971). Putting numbers to the stabilization brought about by each structural feature—+1 unit for each A-U pair, +2 for G-C and 0 for G-U, and various values from -2 to -7 for hair-pin loops, looped out bases in one strand of duplex, and "bulges" of unpaired bases in both strands (these numbers being derived from melting data on models, processed according to straightforward statistical mechanical arguments)—they are able to estimate the relative stabilities of different pairing schemes for a given sequence. Some of the data are still tentative, and will no doubt be improved as more apposite model compounds are synthesized and examined, but it is

Detecting Proviruses

If the RNA tumour viruses replicate by making with reverse transcriptase a DNA provirus—a DNA copy of the RNA viral genome—and if this provirus is integrated into the chromosomes of infected cells, it should, in theory at least, be possible to detect a provirus by DNA/RNA hybridization experiments. Radioactively labelled viral RNA should hybridize specifically with the proviral DNA of an infected cell. There are, of course, enormous technical difficulties, most of which stem from the fact that the proviral DNA must constitute only a minute fraction of the total DNA in an infected cell, and the past years have witnessed a series of claims and counter-claims; the optimists have claimed to detect such specific hybridization and the pessimists have argued the hybridization detected is not specific. But as the report by Baluda and Markham in next Wednesday's *Nature New Biology* suggests, the pendulum is decidedly swinging in favour of the optimists.

Baluda and his colleagues have repeatedly detected some four to ten-fold greater hybridization between the DNA of transformed chick cells and the RNA of the transforming virus—either avian myeloblastosis virus or avian sarcoma virus—than between mouse fibroblast DNA and the RNA of these avian tumour viruses. It seems therefore that transformed chick cells have DNA sequences complementary to the transforming virus's RNA. But the provirus hypothesis demands more than that. If the DNA provirus is to act as a template from which progeny

RNA molecules, destined to become the genomes of progeny virus particles, can be synthesized, then the provirus must contain the entire viral genome.

How can one test whether the viral RNA which hybridizes with a fraction, the putative provirus, of the DNA of a transformed cell comprises the whole virus genome rather than a few regions of that genome? The obvious answer is to recover from the DNA/RNA hybrid the viral RNA and characterize it. But this presents a fresh set of technical difficulties, for it is crucially important not only to recover all the RNA which has specifically and completely hybridized to the cellular DNA, but also to exclude the RNA which has only partially and non-specifically hybridized to the DNA. Faced with these problems Baluda and Markham first fragmented the viral RNA to small pieces, then they hybridized these to DNA of cells transformed by the particular virus, washed the hybrids with saline detergent solutions to eliminate partial hybrids and finally recovered the RNA fragments which had specifically hybridized to the DNA.

To show that this collection of RNA fragments includes the entire viral genome, Baluda and Markham compared the average base composition of intact viral RNA genomes with that of the fragments of RNA recovered from the hybrids. The two compositions are almost identical. The result is therefore at least consistent with the notion, although it does not prove it, that the provirus is a DNA copy of the complete tumour virus RNA genome.

already possible to rule out various of the published models of 5S RNA, for example, with some conviction. Tinoco *et al.* demonstrate what has been so mulishly ignored in the past, that base-pairing can actually destabilize a structure, if it is bought at the cost of too many entropically unfavourable loops and bulges.

There is every promise that this rational approach will prosper. Meanwhile, in a world apart, X-ray crystallographers are pursuing structures, at least of tRNAs, directly. Kim *et al.* (*Proc. US Nat. Acad. Sci.*, **68**, 841; 1971) have reached the point of showing, by virtue of the low symmetry of short segments of base-pairs, which allows the helix spacings to be apparent when viewed from one angle but not from another, that yeast phenylalanine tRNA contains short duplexes, of no more than some 4-7 base-pairs, more or less as expected from the cloverleaf model.

microbiological problems associated with different aquatic environments in and around power stations, including the bacteria of demineralized water in cable cooling circuits, condensate associated with turbine lubricating oils and condenser slimes. Some influences of power station discharge on man-made Lake Trawsfynydd in North Wales were outlined. Mr J. W. Whitehouse and Mrs S. Luff (CERL) on plankton showed that, whereas the phytoplankton are evenly distributed throughout the lake and had shown no changes which might be attributed to warming, the zooplankton exists as two discrete populations, one in the warm lagoon, the other in the larger, cool lake. The two populations of *Cyclops abyssorum* do not differ significantly in size or fecundity. Benthic surveys by Dr D. R. Rothwell (Liverpool Polytechnic) have revealed a lack of fauna in the warm lagoon, but this lack is probably not the result of the raised temperature; the possibility that chelation of trace metals

on peat and erosion is involved is, however, being considered.

A good example of an experimental study being used to aid interpretation of field results was provided by Dr R. J. Aston (CERL), who dealt with the effects of temperature and oxygen concentration on two tubificid worms. Mr A. Aitken (University of Nottingham) showed how lipid can be utilized in place of protein when rainbow trout are maintained at elevated temperatures. The importance of considering fluctuating temperatures as well as constant conditions in experimental studies was stressed by Mr B. G. Lewis (CERL), who showed how, in *Cyclops abyssorum*, variations in temperature can affect the rate of development differently.

Two aspects of work described by Dr K. U. Clarke (University of Nottingham) on the effect of temperature and photoperiod on the endocrine system and chromosome puffing in *Chironomus* sp. are of particular relevance to the identification of thermal effects: tem-

FRESHWATER BIOLOGY

Thermal Effects

from a Correspondent

REPRESENTATIVES from a wide range of institutions attended a symposium on freshwater biology and electrical power generation at the Central Electricity Research Laboratories (CERL), Leatherhead, on April 22. The principal object of this symposium was to present recent CERL work in this field to a wider audience and to give other biologists the opportunity to discuss it.

The first speakers considered the discharge of warmed water from power stations into rivers. In his discussion on the effects of heating polluted water, Mr J. S. Alabaster (Water Pollution Research Laboratory) said that on parts of the River Trent where cooling towers are used, the net effect on water quality and on the status of the fishery seems to be beneficial; water gains oxygen and loses ammonia on passage through the towers. Mr T. E. Langford (CERL) has been unable to show any apparent correlation between the general composition of river faunas and maximum temperature, at least up to 32° C, after sampling around twenty power stations on twelve rivers.

Several speakers expressed the view that biologists did not often provide information in a form which could be readily used by management. For example, there had been some reluctance to classify rivers on some biologically comparative basis. A need for more biological monitoring of rivers was emphasized, and other speakers felt that existing information needed to be collated and codified at once.

Dr J. E. Rippon (CERL) discussed

Morphology of Polyethylene

PRUD'HOMME and Stein report in next Monday's *Nature Physical Science* that they have obtained conclusive evidence of the microspherulitic structure of polyethylene using a novel extension of the techniques of low angle light scattering to polymer films.

Light scattering photographs were taken of films 25 to 725 μm thick using H_v polarization with a laser source, and they exhibited a pattern characteristic of scattering from anisotropic spheres. The diameters of the scattering entities were in good agreement with those obtained for spherulites by Booth and Hay (*Nature*, **227**, 701; 1970) in thin films of low density 'Rigidex' polyethylene; the lower diameters observed in high density fractions and resolvable only with a stereoscan electron microscope suggest that variations in sample characteristics and crystallization procedures can be effective in changing the nucleation density. Nevertheless, it now seems well established that the lack of resolution of the spherulites by light microscopy is caused by sample thickness and in particular clouding of the contours of the spherulites by the large percentage of birefringent units present.

There is, however, no evidence as yet that polyethylene is capable of producing larger spherulites with diameters 0.1 to 1.0 mm except after excessive heat treatment. Accordingly, the conclusions of Banks *et al.* (*Nature*, **194**, 542; 1962) that these structures are not representative of polyethylene samples as a whole—because they are most probably artefacts of the heat treat-

ment (degradation or strain produced on thinning the samples)—must still be considered valid.

Spherulites are the form in which one expects high molecular weight substances to crystallize from the melt, and the only unusual characteristic of polyethylene spherulites seems to be their small size. There is clearly a high nucleation density which would also account for the extreme difficulty in obtaining polyethylene in a quenched amorphous state.

Spherulites of polymeric materials are formed from radiating arrays of fibre-like crystals which have developed from single crystal nuclei by a process of preferential growth along one crystallographic axis and repeated branching. They thus accumulate into sheaves and finally into spherulites.

The final stage of spherical growth is amenable to kinetic analysis in terms of the general crystallization rate equation, $1 - X_t = \exp(-Zt^n)$, which relates the crystallinity to time and in which n is 3 or 4. Analysis of the crystallization isotherms of polyethylene by means of this expression has not been particularly successful because it yields anomalous fractional n values. Because the final morphology of bulk crystallized polyethylene is microspherulitic, the assumption that spherical growth predominates during most of the crystallization process cannot be justified. Further detailed analysis of the fine structure of polyethylene spherulites during their development is clearly required in order to determine a more appropriate rate expression.

perature causes obvious differences in the appearance of adults which can be used as guides to the "temperature history" of the pre-adult stages and the size of chromosome puffs produced can be correlated with the temperature at which the animal is kept.

Mr. I. R. H. Allan (Ministry of Agriculture, Fisheries and Food) chaired the final discussion on future developments. On the research side, Dr J. C. Peters (Water Resources Board) felt that more use would be made of ecological modelling techniques. Mr M. J. Bulleid (Central Electricity Generating Board) thought that the future prospects for using warm discharges in fish farming were good, and on the recreational side the stocking of lakes and rivers for fishing would increase. Several speakers emphasized the need for more work on fish behaviour in relation to power station intakes, particularly in estuaries.

This symposium was useful in showing the wide range of ecological and experimental approaches currently being used to study thermal effects and in bringing together the varied interests involved with water resources. Even if some differences in approach were not exactly resolved, the meeting certainly helped to crystallize them.

CONGENITAL MALFORMATIONS

Teratologists United

from a Correspondent

THE European Teratology Society was formed recently to promote research into the causes, prevention and management of congenital malformations. Its first meeting in University College, Cardiff, on April 14-16 included symposia on all three facets and attracted an attendance of 250. The presence of clinicians, epidemiologists and experimental scientists at the same meeting was very encouraging, because teratology has been separated for too long into its component disciplines. If the society can keep the support of these various groups, it should make an important contribution to the quality of research in this area.

A symposium on the aetiology of human birth defects was addressed by a geneticist (Professor W. Lenz, Münster University), an epidemiologist (Dr I. Leck, University of Manchester) and an experimental teratologist (Dr J. G. Wilson, University of Cincinnati). The issues were spelled out clearly, but little new emerged. There is still almost nothing known about the causes of human congenital malformations, except for a small number attributable to known genetic or environmental factors. It is generally agreed that a genetic susceptibility plus some environmental

triggers are necessary, but the relative importance of these two components is not known. Nor is it known what the relevant environmental stimuli might be, although nutritional factors are indicated by a number of lines of evidence. One new and interesting correlation, reported by Professor C. R. Lowe (University College, Cardiff), is between the softness of drinking water supplies and the incidence of central nervous system defects.

The second symposium examined the problems of screening drugs for potential teratogenic properties. The present procedures were outlined by Professor H. Tuchmann-Duplessis (University of Paris) and were then discussed from the points of view of the pharmaceutical industry (by Dr J. Grauwiler, Sandoz Ltd, Basle) and of the university research department (by Professor F. Beck, London Hospital Medical College). The session concluded with a lengthy discussion. There seemed to be general agreement that current screening methods reduce the danger of another thalidomide disaster, although

some speakers clearly thought that the present regulations are over-cautious and are unnecessarily banning some valuable drugs. The variability of teratogenic response shown by different species was also stressed, and it was hotly contested whether testing in rats, mice and rabbits is adequate or whether primates should also be used. Advocates of using monkeys stress the similarity of the defects induced by thalidomide in man and the rhesus monkey and that rodents are refractory to the effects of this drug. But many research workers remain unconvinced by these arguments and point out that amethopterin is teratogenic in rodents and in human beings but is apparently not so in the rhesus monkey.

It is hard to see how this question will be settled, because the list of known human teratogens is small, and hopefully will remain so. It would seem wise to investigate the possible use of a non-rodent species in testing for teratogenesis, but at present there seems to be no compelling reason why primates should be the best choice.

Morphine and Neurotransmitter Release

MORPHINE is thought to act in the brain by reducing the release of acetylcholine (ACh) at cholinergic nerve terminals. An alternative mode of action could be a reduction of the sensitivity of the post-synaptic ACh receptors by occupation by the narcotic. The potency of morphine-like analgesics is known to be correlated with their potency in reducing ACh released at muscarinic sites in the gut. The more accessible neuromuscular (nicotinic) junction has been used extensively to study quantitatively cholinergic transmission and as a model for studying the action of central nervous system depressants.

In next Wednesday's *Nature New Biology*, Pinsky and Frederickson report that both morphine and its analgesic-antagonist, nalorphine, block supramaximally stimulated transmission at amphibian and mammalian neuromuscular junctions. Nalorphine was the more potent in reducing the nerve-induced twitch of amphibian sartorius muscle and it augmented a morphine-induced depression. The blocking action was increased by d-tubocurarine (d-TC) which competes with ACh at the receptors. Both drugs were effective in concentrations five hundred times greater than that known not to depress nerve conduction and had no effect, at ten times the neuromuscular blocking dose, on directly stimulated muscles denervated three weeks previously.

Recording from single end-plates of muscles paralysed with d-TC, the first small dose of morphine increased the end-plate potential (EPP), but subse-

quent higher doses produced a graded depression of EPPs, again augmented by nalorphine which alone was also depressant. Because high concentrations of morphine are not curare-like on the frog rectus abdominis muscle, neuromuscular block is probably induced by impairing ACh release.

Similar effects were seen on the twitch response of the rat hemidiaphragm to phrenic nerve activity. The initial potentiation of the twitch occurred with lower concentrations of nalorphine and was more prominent and prolonged than with morphine. An anticholinesterase action is unlikely because morphine is more effective than nalorphine as an inhibitor of ACh esterase in the rat brain.

In a second article Frederickson and Pinsky report that morphine reversibly depressed ACh output during indirect tetanic stimulation of the frog muscle, and this narcotic also produced a leftward parallel shift of twitch and of tetanic fusion tension plotted against log (ACh release). This facilitation which occurred even when high concentrations of neostigmine were present suggests that morphine enhances the action of ACh at its receptors. Whether post-synaptic facilitation occurs at cholinergic synapses in the brain and could account for morphine withdrawal symptoms remains of course unanswered, but the authors intend to study other narcotics and their antagonists at the neuromuscular junction as a possible model of what may apply in the brain.

Social Values and Research in Human Embryology

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New techniques for interfering with the early stages of human development raise difficult social and legal issues, which are discussed here by an embryologist and a lawyer. This article is based on a lecture and discussion held at the National Law Center of the George Washington University in December 1968.

UNTIL recently, there have been few scientific studies on human conception and early embryonic growth, partly because the eggs and embryos were unavailable, and partly because various laws reflecting certain social attitudes towards human life discouraged change. The common law of homicide of early centuries dealt only with entire humans, either the mother or fully delivered child¹. Legislation during the nineteenth century made abortion a crime against the mother², and then against the foetus *in vivo*³. The latitude given by case law since 1939 for aborting the victim of rape⁴ was scarcely a mandate for widespread abortions in the interests of married couples. Only in 1967 did the English statute law take notice of what may be called "eugenic abortions", done not in defence of the life or health of the mother, but in defence of the family and indeed of society⁵. (As of mid-1970 more than half of the States of the USA still forbade eugenic abortions.) These changes in abortion law have so far taken place years after the event that precipitated them. The increasing tempo of scientific achievement now seems to demand that social attitudes are helped to keep pace with or even direct new issues as they arise.

Even today intervention into foetal development is limited to methods such as artificial insemination, the abortion of unwanted fetuses at three months of gestation and later, or averting the birth of rhesus babies by transfusing the fetuses at great risk. These issues have already raised controversy; the intense and continuing debate over legalized abortion has shown the depth of public feeling on issues affecting human reproduction. The stimulus for this article is close at hand⁶: the rapidly growing ability to fertilize human eggs in culture, grow them for three to four days in the laboratory and then replace them in the mother to grow to full term. The steps in this basic procedure are becoming well understood. The wife is stimulated with hormones to produce several ova in a menstrual cycle. By means of minor surgery under general anaesthesia one or more ova are withdrawn through the abdominal wall, a procedure that can be done repeatedly. The ova are then fertilized with the husband's sperm, and within five days or so they have grown in the laboratory to more than thirty-two cells. The last step to be taken when more is known about the embryonic development will be to replace the embryo in the wife so that it will implant and grow to full term to be delivered naturally.

The procedures leading to replacement and implantation open the way to further work on human embryos in the

laboratory. We believe that it is important at this stage to elaborate the emerging issues in order to give time for defining and evolving social attitudes on which to base rules of conduct for scientists and society. We want to sketch some of the classes of scientific enquiry now under way, on humans and animals, to outline some of the emerging social problems and legal rules in Great Britain and the United States, and to offer comments and suggestions on ways of helping society, science and law to move more swiftly, harmoniously and with greater confidence in keeping pace with advances in human embryology and other disciplines.

Alleviating Infertility

A most immediate application of fertilizing human eggs and growing them *in vitro* is to bypass a blockage in the wife's Fallopian tubes. When the fertilized ovum has grown to eight or sixteen cells, it is probably sufficiently developed for replacement in the mother. These women could thereby have their own children, fathered by their own husbands. There would probably be demand for thousands of babies by this means every year—babies that are not born now.

The social issues that emerge involve health, economics and law. The physical health of the parents does not demand that their infertility be cured. Some couples may find adoption to be a satisfactory answer to the problem, which would also relieve social and personal problems of children without parents. Yet the desire to have children must be among the most basic of human instincts, and denying it can lead to considerable psychological and social difficulties. Moreover, it is no longer certain that children will be available for adoption as abortion becomes more acceptable.

Infertility seems to be a clinical defect to be remedied if possible by medical attention. It cannot be a rational basis for denying on ethical or other grounds the right of some couples to have children. There is no particular genetic reason to suppose that the handicapped mothers will have children with similar handicaps. Granting that a previously infertile couple should have two children if they want them (by whose permission—their own? their physician's? demographers? bureaucrats?) "should" they have four, five or ten? Several hundreds or thousands more babies might be born every year in Britain if embryo transfer were available. Social concern for overpopulation is now widespread. Our view is that a campaign against overpopulation should be directed to all parents generally and not enforced on a selected few; it would scarcely seem a justifiable public policy to prevent such couples from having their own children.

Is there any guide in law about the growth of human pre-implantation embryos outside the body, and especially about the disposal of those not replaced in the mother? The legislators that wrote the anti-abortion statutes, not foreseeing the creation of unimplanted blastocysts in culture, provided criminal penalties only for destroying or attempting to destroy implanted fetuses. Under the Offences against the Person Act², for example, the operative words are "Whosoever, with intent to procure the miscarriage of any woman, whether she be or be not with child, shall unlawfully . . . use any instru-

ment. . . .” Under the customarily strict rules for construing criminal statutes, it is very doubtful that today’s embryologists and doctors have the requisite criminal intent to defy these statutes or that the death of a blastocyst *in vitro* is a miscarriage.

Does the law of battery apply to the treatment of the mothers? This law comes in two forms, civil (damages) and criminal (fine and/or imprisonment). Consent is an absolute defence to civil battery, that is touching another without consent, and the mother involved in studies or experimental embryology certainly consents. Whether she or anyone else can consent on behalf of the foetus is highly speculative. Criminal battery is prosecuted by the government in Britain. Yet it seems doubtful that battery, usually a trifling offence, is relevant to the creation or implantation of the foetus.

Determining Sex of Embryos

The basic procedure of embryo transfer will probably be combined with the identification of the sex of the embryo. Sexing can now be done with complete accuracy in the rabbit⁷, although the procedures are far from perfect. The eugenic reason for sexing is that many genetic disorders are sex-linked, hence they usually occur in males. In couples with a history of genetic disorders, only female blastocysts will be replaced in the wife. Some daughters born through this form of genetic selection will be carriers of defective genes, and the selection process will have to be repeated with the embryos of these daughters in subsequent generations. At present, male foetuses thought to have sex-linked disorders can apparently be legally aborted under British statute. Under the Abortion Act⁵ an abortion can be performed under specified safeguards where “there is a substantial risk that if the child were born it would suffer such physical or mental abnormalities as to be seriously handicapped”.

Even if the law was interpreted to say that destroying a blastocyst is abortion, the opportunity to give the mother a healthy embryo seems fundamentally humane. Sex-linked defects are only one sort of problem that might be avoided by this procedure; chromosome examination may one day prevent the birth of individuals with genetic anomalies such as mongolism. But the law in most places apparently is simply not ready for this sort of procedure. For example, the availability of ordinary means of lifesaving medical care can create a legal duty on parents to avoid jeopardizing an unborn infant’s life. In New Jersey a mother can be forced to accept a blood transfusion to save her baby’s life before delivery⁸. Yet to abort a foetus for a congenital defect would apparently be a crime in the same State, as members of the same court pointed out three years later in denying damages for medical malpractice to a deformed baby born after his mother contracted German measles during pregnancy⁹. No other American State has dealt with this sort of problem in comparable detail.

A major social issue arising through sex determination could be demographic imbalance between the sexes. The selection of girls for purely eugenic reasons must take a long time to build into statistically significant proportions, if ever. But how about giving parents the opportunity to choose boys or girls, for example in providing a boy for the five-girl family? No one knows what the consequences might be, although they could be serious if there was a significant shift in the sex ratio. The free choice of sex of offspring, even if widely available almost at will of the parents, might have no more identifiable impact on population than giving women the vote had on voting patterns. Various forecasts have suggested that given free choice there would be a con-excess of boys; others question this¹⁰. But even a small shift to boys would be no light matter, for the natural excess of male births in some populations is now persisting into older generations, and any bias towards pre-selecting boys would thus exacerbate this situation with its consequences of a “raiding” of the younger age-groups of women by older men.

Modifying Embryos

Further developments that also depend on the growth of early human embryos perhaps contain the most controversial issues. These developments involve not simply identifying sex or genetic defects, but rather modifying or adding to the embryo itself. Two emerging possibilities are especially relevant to this time. First, donor cells can be injected into a mouse embryo and their multiplication during foetal growth can lead to the partial colonization of many organs. The foetus thus grows as a “chimaera” with its own characteristics modified by those of the donor¹¹. It now seems that the various body tissues of an adult each grow from their own particular small pool of cells formed early in development—possibly even when the embryo can still be transferred into the mother¹². For example, fewer than half a dozen cells are reputed to be progenitors for the blood forming tissues in the human body¹³. Obviously, a few cells injected into the embryo at this critical time could profoundly influence development. Precursor cells that are specific for particular tissues are not yet available, and donor cells from other blastocysts would have to be used.

Benefits stemming from this treatment could include “masking” various genetic diseases in the recipient, but the donated cells would also partially colonize brain and nervous tissue. More distantly, donor cells or the whole embryo might be modified using nucleic acids as carriers of genetic information. While a case might be made for using the method to mask genetic deficiencies, the treatment would not necessarily be limited to this aim. Selection of the appropriate precursor cells could be used one day profoundly to influence personal characteristics. In effect, a composite organ transplant could be offered to an offspring while it was a pre-implantation embryo. Already, newborn children suffering from deficiency diseases have been given donor cells capable of repairing the deficiency (ref. 14; thymus grafts have also been given to neonates). Colonization during the earliest stages of development would obviously increase the chances of close relationships between host and donor cells.

The second possibility is more extreme, although more remote because it is technically difficult to accomplish. Experimental work in animals has shown that the chromosomes or nucleus can be removed from an ovum and replaced with a nucleus taken from a cell of a donor. The embryo would then grow displaying characteristics of the donor rather than its own. Nuclear transfer can be done manipulatively in amphibian eggs, and by means of viruses to fuse a donor cell with a mouse egg*. There is no need to stop at one egg—several or several hundred eggs could be “cloned” in this manner and might all develop into identical or closely similar offspring.

What advantages and disadvantages would arise from cloning? Comparisons between cloned children and identical twins are of limited value, for twins are born together and grow up together. Cloned children would be closely similar to an older, earlier individual, that is the nuclear donor, and therefore they would have been deprived of what would be interpreted as one fundamental human right, the right to be different. The cloned person could be intelligent, motivated, highly adapted to a technical society, just like the successful donors. He would probably be given a good education, be physically strong, and very successful. Such offspring could be so well adjusted and distinguished that any decision not to produce them could be strongly criticized. But it is equally possible that a child composed by nuclear transfers would be a psychological misfit. He might become infatuated with the donor’s background, and would be virtually unrelated to his familial parents. There would be a strong impulse to treat cloned children as laboratory subjects to be analysed repeatedly during their lifetime. Standardization of the genotype of many foetuses by nuclear transfer could provide major insight into the effect of

* In *Xenopus laevis* the transferred nucleus seems to dominate the characteristics of the offspring¹⁵. Experiments with mammalian ova are described in ref. 16.

environment on such intricate traits as personality, adaptability, and so on, but at what cost? Adoption of this method of analysing human traits would raise a much more difficult question: who sets up the criteria for establishing these "models", decides where they develop, and determines their environment?

The social issues in the reactions of offspring from cloned, chimaeric or "sexed" embryos could have parallels with the psychological reactions of children on finding out that they are adopted, and of organ recipients, especially of hearts¹⁷. Sexing could probably be justified on these grounds, but extensive carbon-copying of genetic models of people, or chimaerism, should be questioned strenuously, in spite of the benefits claimed for some of these procedures*. It is highly doubtful that scientists or doctors could know in advance of any social benefits arising from these methods when we have so little information about genetic and other factors that influence complex human characteristics. It is also questionable that the donor's characteristics would be totally inherited in the offspring, since even small mutations in the donor's cells or uterine effects might have subtle effects on the offspring. In our opinion, the objections and doubts about cloning, let alone the current technical difficulties or the problem of who is worth copying, dismiss any consideration of using this method at present. Does each child have a human right to be an individual, and is there a correlated human duty resting on scientists not to create human clones or chimaeras? This right is not the right to life covered in parliamentary debates on the Abortion Act; this is the right to be one's self.

The law is again unprepared to deal with such complex questions of paternity. An easy but merciless response in artificial insemination (donor) cases is to declare the child illegitimate²². Even so, the parents could acknowledge or adopt and thereby confer the full rights of inheritance on the child. Acknowledgment (of paternity) places the bastard child on the same footing as regards inheritance or other property rights as legitimate children, the methods ranging in different American States from conduct by the father towards the child to judicial proceedings. It would be hard to advise parents of cloned offspring because of ambiguity concerning whether the child is related to the parents (acknowledge) or unrelated (adopt).

Regulating Applications

Would the new human embryological techniques threaten major social values, such as the sanctity of life, the independence of scientific inquiry, or service of the best interests of patients by medicine (see refs. 18–21)? It is difficult to say how much the sanctity of human life has already been impaired (if at all), in the eyes of people generally, by reducing the inevitability of birth through methods of contraception and abortion. Eugenic techniques providing healthy births should enhance rather than harm the sanctity of life, and science and social progress would move together humanely, as they appear to be doing through moves and laws on contraception, abortion and homosexuality. Nevertheless, there have been occasional criminal statutes and common law decisions which judged that doctors and scientists working in areas of inquiry found to be especially sensitive were not to be left alone. In rare cases, for example, body snatching for anatomical dissection, the work was forbidden²³, although more often it is channelled according to social rather than scientific objectives as in the use of new drugs or contraceptives†. Adverse social judgments have not diverted dedicated doctors and scientists convinced of the value of their work: for example, family planning passed through this stage yet is now widely accepted.

Does anything need to be done to regulate the application

* These are discussed in *Literaturnaya Gazeta* by Lederberg and Badalyan¹⁸. Later, Lederberg made no reference to cloning and discussed the potential importance of molecular biology on research into human developmental physiology¹⁹. Doubts about the overall success of cloning are discussed in ref. 20. See also ref. 21.

of new scientific and clinical advances?—Why not *laissez-faire*? Almost any scientific advance can be used for good or harm, and free and open discussion of emerging issues—an invaluable safeguard—is readily available nowadays. (Some scientific or ethical aspects of work on human conception are discussed in ref. 27.) But communication is distinct from regulation. Well developed governmental and private systems of discipline and compensation already exist, and they could be brought to bear at every stage of scientific advance in human reproductive studies. The difficulty with all of them—civil and criminal sanctions, the proceedings of professional organizations, and government regulations—is that they all contain an implicit direction, "Ask (someone's) permission before you do your research". Scientific research began to make progress only when it no longer had to ask permission of the church, and it has come a long way since then. Yet the scientist's freedom to inquire is not immutable; society might again force scientists to consult institutions that were not developed for judging the motives of humanitarian biologists and physicians. People are bound to take an interest in what others propose to do with their successors, even in the name of scientific inquiry. What is to be feared is that if the biologists do not invent a method of taking counsel of mankind, society will thrust its advice on biologists and other scientists and probably in a manner or form seriously hampering to science.

Probably the worst consequence imaginable to scientists working in a political democracy would be the pre-emption by the state of a branch of science such as human embryology. Pre-emption is not unprecedented, where the need seems acute enough or nationalization is the rule, as with nuclear weapons or chemical and biological warfare. But in a hitherto free science, confining research into a government agency would tend to stifle inquiry and might promote secrecy, whereas full information and free discussion seem to be the best possible antidote to public concern. Alternatively, the state might decree a temporary moratorium on embryological research on humans. By analogy, prohibition of alcoholic beverages might be a good idea, but neither prohibition nor moratorium seems likely to be achieved in the real world, because in both cases the work is easy to do and to conceal, and if one nation prohibits it, others will permit it, leading either to smuggling of the work or emigration of the people who do it. Statute laws in scientific areas, once passed, are hard to change, and they may be a continuing menace to those engaged in later work that is quite acceptable to the community.

Forms of regulations or consultation intermediate between *laissez-faire* and state pre-emption might be useful. In many professions various forms of self-regulation exist already, and could be adapted to meet present needs. In theory, judges regulate lawyers through the courts; in application, lawyers regulate themselves through disciplinary committees with judges presiding, although in practice very little regulation takes place except in cases of misbehaviour such as stealing from clients. In contrast with many other professions scientists largely regulate themselves at present and the few regulations that do exist, such as the Home Office rules on animal experiments, are concerned with form rather than the content of the work. Delegating regulations to individual physicians or medical organizations is an attractive possibility, for they will be called upon to apply new techniques to their patients. The beginning of medical ethics, however, is *primum non nocere*; this permits alleviation of infertility, and has been stretched to cover destruction of foetuses with hereditary defects, but would it permit the more remote techniques like modifying embryos? Moreover, individual doctors have placed what they believe

† One example is the consent of patients before they are treated with investigational pharmaceuticals²⁴, another is forbidding the use of contraceptives, which was declared unconstitutional in the United States²⁵; forbidding the advertising, sale or giving away of contraceptives is now under debate, although statutes so forbidding are common, and the US Supreme Court agreed in March 1971 to hear a case on gifts of contraceptives²⁶.

to be the interests of their patient before those decreed by society, for example those abortions carried out in the face of severely repressive laws. The physician is the necessary link between biologists and the patient or those persons who want to act as subjects or donors in experimental work, and this is a valuable control. But medical ethics perhaps look best when compared with the absence of organized self-regulation in other occupations and undoubtedly operate best with medical problems. It is doubtful if they are superior to any other system when wider issues are at stake, for example questions of abortion, or on whether to go ahead with embryological experiments. The institution of medicine is a closed shop and therefore represents one set of opinions; physicians have a large say already in experimental human biology and there seems little reason to give them the only voice.

Another relevant model of intermediary regulation is research in pharmaceuticals. The basic invention and development and testing in lower animals are done predominantly by non-physicians (pharmacologists), the government steps in to regulate experimentation on humans by physicians and the accumulation of statistics by pharmacologists, and eventual marketing and monitoring are again controlled by governmental or quasi-governmental organization acting in concert with physicians or drug firms. Without pursuing parallels and differences in detail, it would seem that if the least government is the best government, this would be the best model for state regulation. It is doubtful whether this model could be applied to cover the diverse techniques developed in different laboratories during research. Nor has it been practised in many areas of clinical advance where new surgical methods or other novel approaches were under investigation. Nor does it seem that conclusions drawn from animal embryos would necessarily apply to man, for there is obviously no parallel in animals with our intricate knowledge of human emotions and behaviour. Moreover, scientists would probably prefer a method that leaves government out of the picture altogether, and a method that gives them assistance in whether to inquire or not. Scientists must maintain their right to exercise their professional activities to the limit that is tolerable by society in order to develop new concepts or discover new facts that could be of great value to our health or understanding.

If some form of regulation is required, perhaps what is needed is not heavy handed public statute, or rule-making committees, or the conscience of individual doctors, but a simple organization easily approached and consulted to advise and assist biologists and others to reach their own decisions. Such an organization must represent widespread but uncommitted interests and be free of partisan politics. It would frame public debate, act as a watchdog, and yet interfere minimally with the independence of science. Decisions need to be authoritative yet challengeable, debate easy and constructive, dissension emphasized and judgments respected rather than enforced. Some of these characteristics are to be found in existing institutions; for example the Press Council or the Ombudsman*. In line with the stress on individual scientific responsibility, advisers are needed whose achievements and attitudes make them worth listening to in matters affecting the future of human research—doctors, scientists, lawyers, authors, and other laymen—because they are broadly talented people, not because they serve *ex officio* as formal representatives of their professions, societies or constituencies. Such an erudite source of opinion and reliable information would have great value for the press, teachers, theologians and others interested in making up their own minds or influencing others. Rulings should be as altruistic and detached as possible, powers of subpoena, funding research, or veto over particular work would be avoided as inconsistent with the advisory role. The

authority of such an organization would not be negligible: a funding committee for example, with doubts about the human implications of a proposed research project, might well seek advice by sending applications to it for an opinion.

The stress would be on individual and private action, inquiry and consultation, not on authority, control bureaucracy or "laws with teeth". A test period of two or three years for such an organization would do no harm, as long as it is not expected to do the impossible. It would probably take a fairly conservative line as lay attitudes struggle to catch up with what the scientists can do. The organization might die from inaction, unless given a wide scope, for the number of investigations requiring imperative social analysis is very small in comparison with the number in progress. If the group failed, something might well have been learned. The other models involving more or less governmental intervention will still be available, and considering the difficulty of removing governments from a field they once occupy, scientists will at least have the satisfaction that they tried in effect to regulate their own affairs.

Would any scientist use such an advisory group? One could only suppose that they would, if only for their own protection; doctors would probably rely on their own organizations. Most scientists are concerned with human values in their work, although some have not shrunk from secret work with damaging implications for mankind, such as germ warfare and nuclear weaponry. Stimulating public debate on such secret work would be an invaluable social service. With non-secret work, possibly only scientists themselves can effectively regulate their research, and then only within fairly wide limits that include a margin for error as well as differences of opinion.

Time for Action

In human embryology as in other areas of swift scientific advance, the achievements of science catch unprepared a society that lacks either ready made attitudes or the institutional means of forming new ones. A conventional social response nowadays to some novel threat to stability is to demand formulation of law to regulate the development and application of new achievements. But few scientists have any lawmaking experience, and hence their judgments will be those of citizens giving their personal social views. Neither do the lawyers have much of an advantage in creating good social policies toward scientific achievements: legislation and policy-making are not stressed in British or American legal education, lawyers as a group feel little professional responsibility to define public policy and litigation is likely to come too late and be too haphazard to effect major social policies in a short time.

When scientists clearly foresee potential conflicts with existing rules of society arising from their work, paradoxically both human progress and scientific freedom may hang on their activism in arenas generally regarded as social or political. Scientists may have to make disclosures of their work and its consequences that run against their immediate interests; they may have to stir up public opinion, even lobby for laws before legislatures, in the hope that the attitudes of society as evidenced in its laws will mature at a rate not too far behind the transition of scientific discovery into technological achievement. The lobbyists for reform in the laws of drug and alcohol addiction, abortion, and sexual behaviour have achieved much public approval in their areas of concern; can biologists in experimental embryology expect so much more by doing any less?^{2,3}

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² The statute law apparently originated in Act of 43 George III, Chap. 58, Sections 1 and 2 (1803); its substance now in force is the Offences against the Person Act 1861, 58.

³ Infant Life (Preservation) Act 1929, 1.

⁴ The King v. Bourne, 1939 1 K.B. 687 (1938).

⁵ Abortion Act, 1967, 1.

* A commission on Civil Rights was established in the United States in 1967, and is concerned with the deprivation of Civil Rights such as the vote or equal protection under the law. In the United Kingdom the Protection of Human Rights Bill was read for the second time in the House of Commons on April 2, 1971 (ref. 28).

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Drug Receptors and their Function

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There have been various theories about the way in which drugs exert their effects. If recent progress in the isolation of receptors continues, it should soon be possible to look directly at the interaction between drugs and tissues.

THE ability of many cells to respond in a highly selective way to minute concentrations of particular chemicals provides the starting point for the study of pharmacology. Three striking characteristics of drug action have led to the hypothesis that cells possess specific receptors for drug molecules; these are high potency (drugs that act at around 10^{-9} M are common, and effects have been reported at concentrations lower than 10^{-11} M), chemical specificity (exemplified by the marked differences in potency observed between optical isomers) and biological specificity (adrenaline, for example, having a marked effect on cardiac muscle, but very little effect on striated muscle). Langley¹ first suggested the existence of specific drug receptors to account for the action of nicotine in causing contraction of voluntary muscle when applied to a particular region of the muscle (corresponding, as we now know, to the region where the motor endplates are situated) and the antagonism of this effect by curare. The idea was elaborated by Hill², who produced an algebraic formulation based on the law of mass action, but it was not until about twenty years later that the work of A. J. Clark^{3,4} led to fairly general acceptance of the receptor theory. Contemporary with this development of ideas about drug receptors were the early studies on the binding of oxygen by haemoglobin, and on enzyme kinetics, leading to the Michaelis-Menten theory. Since these three lines of research of study all involve the binding of small molecules by proteins, it is not surprising that there is a strong formal similarity between the various

theories that have been explored. Nonetheless, it is clear to even the most optimistic pharmacologist that present understanding of drug-receptor mechanisms lags far behind knowledge of enzymes or of haemoglobin.

The principal factor responsible for this is, I think, that it has not yet been possible to isolate any pharmacological receptor, so such knowledge as we possess has had to be gained by frustratingly indirect methods, and none of the more direct chemical approaches that have been so fruitful in the study of enzymes are yet applicable to drug receptors. It seems high time that this technical barrier was overcome.

Several reviews⁵⁻¹¹ and symposia¹²⁻¹⁶ have appeared in recent years, from which a good idea of the principal theories and lines of advance can be obtained. In this article I want to consider three aspects of the drug-receptor problem in more detail. First, the laws governing the binding of drug molecules to receptors; second, the relationship between the binding of drug molecules and the production of a biological response; and third, progress towards the isolation of receptor molecules. To keep the discussion to a manageable size, I will for the most part refer to the action of cholinergic drugs on cell membranes, which has probably been more thoroughly studied than any other type of drug action.

Binding of Drug Molecules

The simplest possible theoretical model for the drug-receptor reaction, which was first explored by A. J. Clark⁴, has proved remarkably durable. It is assumed that the tissue possesses a population of receptors for a particular drug and that these receptors are (a) identical and (b) non-interacting. By "non-interacting" we mean that the binding of a drug molecule to a given receptor has no effect on the probability that a neighbouring receptor will be occupied. As I shall discuss later, this does not necessarily mean that the effect produced by the occupation of a given receptor is independent of whether or not its neighbours are occupied.

If each receptor can bind a single drug molecule (which is

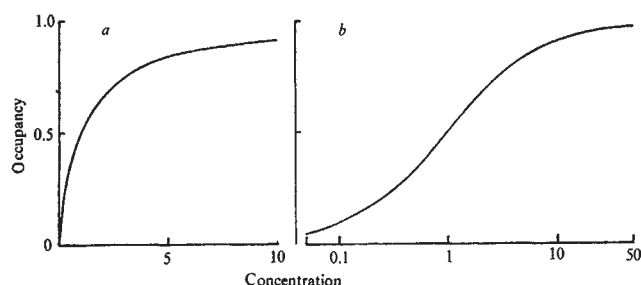


Fig. 1 Relationship between fractional receptor occupancy and drug concentration according to the Langmuir equation. *a*, Plotted on linear coordinates. *b*, Concentration plotted logarithmically. Concentration is expressed as a multiple of the equilibrium constant for the drug.

a matter of definition rather than an assumption) then, from the law of mass action, the fraction of receptors occupied at equilibrium is given simply by the Langmuir equation

$$p_A = \frac{x_A}{x_A + K_A} \quad (1)$$

where p_A is the fraction of the total number of sites occupied by drug A , x_A is the concentration of drug A , and K_A is the dissociation equilibrium constant. This equation corresponds to the familiar hyperbolic saturation curve (Fig. 1*a*) or, when p_A is plotted against $\log_{10} x_A$, to a symmetrical sigmoid curve (Fig. 1*b*) that has a maximum slope at the midpoint of 0.58 per ten-fold change of concentration.

The most obvious, but least satisfactory, test of this hypothesis is to see whether concentration-effect curves for drugs can be fitted by equation (1), on the assumption that the effect produced by a drug is directly proportional to the fraction of receptors occupied. A convenient procedure is to use the Hill plot ($\log [E/(E_{\max} - E)]$ versus $\log$ concentration, where E is the effect produced and $E_{\max}$ is the maximal effect). If $E/E_{\max}$ is proportional to the occupancy, p_A , and equation (1) is obeyed, the Hill plot should be linear and of unit slope. Values for the slope of the Hill plot for various drug effects are collected in Table 1. In most cases, the slope is not equal to unity, so that this evidence cannot be taken to support the use of equation (1) to describe the binding curve. It has been suggested that a modification of equation (1) should be used, namely

$$p_A = \frac{x_A^n}{x_A^n + K_A} \quad (2)$$

The slope of the Hill plot is now equal to n , so the results in Table 1 might be taken to indicate that n may be greater than unity. In some cases, for example, the action of γ -amino-

butyric acid (GABA) in increasing membrane conductance in crustacean muscle¹⁷, n is exactly two, and a mechanistic interpretation of equation (2) is that two drug molecules must combine simultaneously with, and dissociate simultaneously from, the receptors. Where n is non-integral, as is more commonly the case (Table 1), this interpretation is not possible. Recently, however, allosteric mechanisms, analogous to those described for various enzymes¹⁸⁻¹⁹ and for haemoglobin²⁰, have been suggested in relation to drug action^{21,22} (discussed later) and it has been shown that on this hypothesis (which explicitly abandons the assumption of non-interacting receptor sites) the slope of the Hill plot does not have to be an integral number but can assume any value.

As an alternative to postulating a more elaborate binding function than equation (1) to account for the anomalous slopes indicated in Table 1, we can simply assume that the effect measured is not directly proportional to the occupancy, p_A . In many cases this assumption is amply justified on physiological grounds. Thus, where the response measured is contraction of a muscle, the chain of events is probably that the drug causes an increase in membrane permeability, which leads to depolarization of the membrane; this in turn releases calcium intracellularly, which activates the contractile machinery. Now very simple considerations show that the depolarization, ΔV , will not be linearly related to the conductance increase, Δg , produced by the drug²³; and there is also good evidence that the contraction is very far from being simply proportional to ΔV ^{24,25}. To assume that contraction is proportional to receptor occupancy is therefore unlikely to be generally valid and the shape of the concentration-effect curve is correspondingly unhelpful in throwing light on the nature of the concentration-occupancy relationship. The wealth of evidence supporting the existence of "spare receptors"²⁶⁻³⁰ for many contractile tissues responding to substances such as acetylcholine and histamine lends emphasis to this point. The existence of spare receptors means that a maximal response can be obtained when only a small fraction (probably 1% or less for many cholinergic agonists in smooth muscle^{28,29}) are occupied, and is not compatible with a linear relationship between occupancy and effect.

Evidence obtained from the quantitative study of drug antagonists³¹⁻³⁴, on the other hand, tends to support the validity of the Langmuir equation for the binding of both agonists and antagonists. Thus, when the law of mass action is applied to the situation where two drugs, A and B , compete for reversible binding to a single population of receptors, it is found that

$$p_A = \frac{x_A/K_A}{x_A/K_A + x_B/K_B + 1} \quad (3)$$

If agonist concentration x_{A_0} tested on its own produces the same occupancy as concentration x_{AB} tested in the presence of

Table 1 Slopes of Hill Plots at Mid-point of Concentration-Effect Curves for Different Drugs

Drug	Mechanical responses	Response	Slope of Hill plot* at 50% max. response	Reference
Acetylcholine		Guinea-pig ileum, isotonic contraction	2.8	39
		Guinea-pig ileum, isometric contraction	1.9	Rang (unpub.)
Acetylcholine		Frog rectus abdominis, isotonic contraction	0.8	50
Carbachol		Chick biventer cervicis, isometric contraction	3.3	67
Carbachol		Leech muscle, isometric contraction	0.8	75
Acetylcholine		Frog ventricle, inhibition	0.9	50
	Membrane responses			
Carbachol		Eel electroplax, depolarization	1.9	42
Decamethonium		Eel electroplax, depolarization	1.9	42
Phenyl-trimethylammonium		Eel electroplax, depolarization	1.7	42
Diethyl decamethonium†		Frog motor endplate, conductance increase	1.5	Rang (unpub.)
Gamma-aminobutyric acid		Crustacean muscle, conductance increase	2.0	17

* Plot of $\log [E/(E_{\max} - E)]$ against $\log$ concentration. For Langmuir equation slope = 1.0. † 1,10 bis-(dimethylethylammonio)decane.

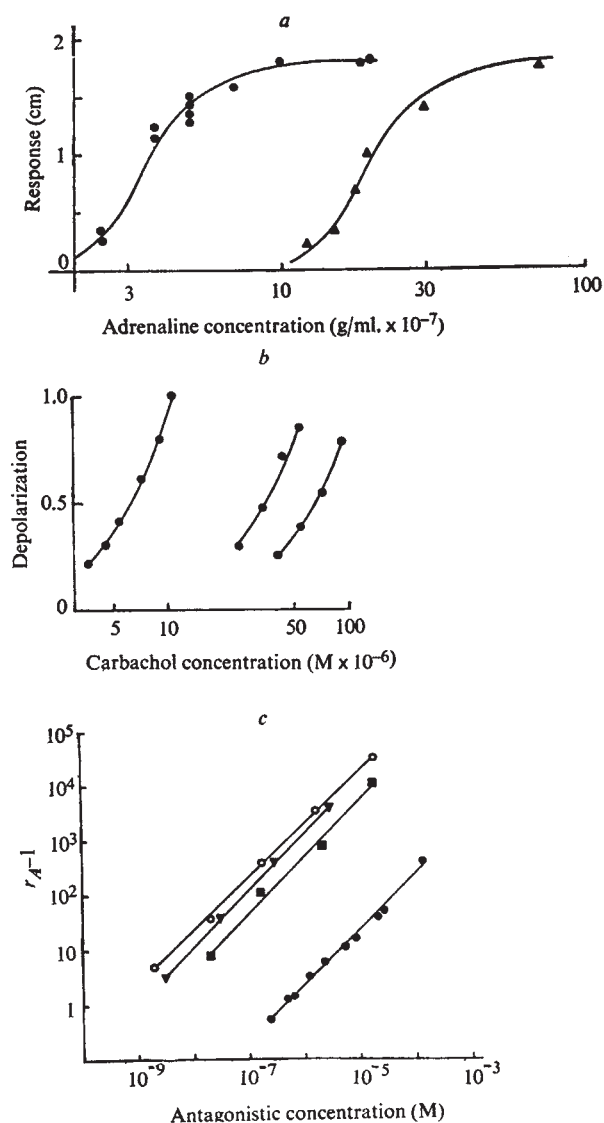


Fig. 2 Drug antagonism. *a*, Antagonism of adrenaline by ergotamine on rabbit uterus (from Gaddum⁷⁶). ●, No antagonist; ▲, ergotamine, 5 µg/ml. *b*, Depolarization of endplate region of frog toe muscle by carbachol, and its antagonism by tubocurarine. Depolarization is expressed in arbitrary units (plotted from results of Jenkinson³³). Left hand curve, no tubocurarine; middle curve, 2×10^{-6} M; right hand curve, 4×10^{-6} M. *c*, Data plotted according to equation (5). ○, Atropine-carbachol on guinea-pig atrium⁵⁵; ▼, atropine-acetylcholine on guinea-pig ileum³²; ■, propranolol-isoprenaline on kitten atrium⁷⁷; ●, tubocurarine-carbachol on frog endplate³³.

concentration x_B of the antagonist, then the ratio, r_A , of these agonist concentrations is given by

$$r_A = \frac{x_{AB}}{x_{A0}} = \frac{x_B}{K_B} + 1 \quad (4)$$

or

$$r_A - 1 = \frac{x_B}{K_B} \quad (5)$$

If the effect produced by *A* is a function of p_A alone, then equation (5) means that the antagonist should simply shift the log-concentration-effect curve for the agonist to the right, without changing its slope or amplitude (this is because r_A depends only on x_B and not on x_{A0}). The magnitude of this shift (that is, the dose ratio, r_A) should be linearly related to the antagonist-concentration, x_B . Both of these predictions have been amply borne out by experiment in many different situations, some examples being shown in Fig. 2. In Fig. 2, logarithmic coordinates are used in testing equation (5), $\log(r_A - 1)$ being plotted against $\log(\text{antagonist concentration})$ ³⁰;

this should give a straight line of unit slope, the abscissal intercept providing an estimate of the equilibrium constant, K_B , for the antagonist. If, however, the binding function for the agonist was correctly represented by equation (2), rather than equation (1), and that for the antagonist was similar, that is

$$p_B = \frac{x_B^m}{x_B^m + K_B} \quad (6)$$

then equation (5) would take the form

$$r_A^n - 1 = \frac{x_B^m}{K_B} \quad (7)$$

and the logarithmic plot of dose ratio against antagonist concentration would be non-linear, having slope m at low dose ratios and slope m/n at high dose ratios. The results shown in Fig. 2 (together with many more experimental examples) thus constitute evidence in favour of the simple Langmuir equation ($m=n=1$) as the binding function for both drugs, although they provide an estimate of the equilibrium constant only for the antagonist. There are very few examples^{16,34} where the agonist-antagonist interaction deviates markedly from this pattern.

Relation between Binding and Effect

It has usually been assumed that the binding of an agonist molecule "induces" a change in the receptor which gives rise indirectly to the drug effect, such as an increase in the ionic permeability of the cell membrane. There is, however, good evidence²⁷⁻²⁹ that drugs which act at the same receptor site can differ in their effectiveness in bringing about this perturbation, since different drugs can produce an equal biological response when occupying different fractions of the receptor pool. This means that the link between the binding and the perturbation of the receptor must be flexible, and this has been characterized by an operational quantity termed "intrinsic activity"³⁵ or "efficacy"²⁷ (efficacy being measured by the reciprocal of the fractional occupancy required to produce a response equal to 50% of the maximum of which the tissue is capable). It has, however, been shown for depolarizing drugs acting on the motor endplate that the reversal potential does not depend on the efficacy of the drug^{36,37}, being the same for a considerable range of substances that have been tested. This finding shows that the ionic selectivity of the permeability channels opened up is the same for all these drugs, and suggests that the "active" state of the receptor is also the same for different drugs. Consequently efficacy is more readily interpreted as a measure of the probability that an occupied receptor will assume the active conformation, than as a gradation of the conductance increase mediated by each individual receptor. The evidence is still far from complete, but tends to favour a mechanism by which an agonist molecule increases the probability of a quantal transition of the receptor from an inactive to an active state.

An extremely interesting recent finding by Katz and Miledi³⁸, which may provide an experimental approach to the problem of the quantal nature of drug action, is that the membrane noise (that is, spontaneous random fluctuations of potential) at the frog motor endplate increases when the membrane is depolarized by acetylcholine (but not when it is depolarized by passing current). Analysing this phenomenon, Katz and Miledi assumed that the depolarization was built up of a series of elementary events each consisting of an instantaneous depolarization of fixed amplitude followed by an exponential recovery with a time constant of 10 ms (an approximation to the time course of passive electrotonic decay in a muscle fibre), as shown in Fig. 3*a*. From the root mean square amplitude of the noise, they calculated that the amplitude of the elementary event was 0.29 µV, which is much too great to represent the movement of a single ion across the membrane. An average depolarization of 10 mV would correspond to a frequency of these elementary events of about $3 \times 10^6 \text{ s}^{-1}$. It

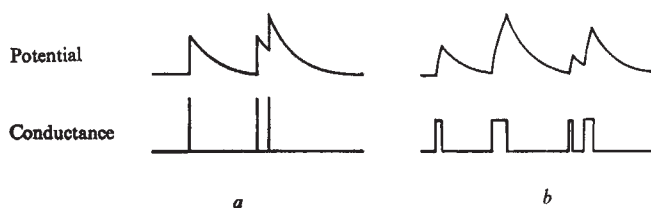
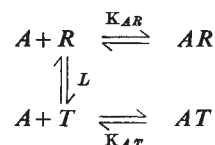


Fig. 3 Possible interpretations of membrane noise produced by depolarizing drugs. *a*, As assumed by Katz and Miledi³⁸. Individual channels open for an infinitesimally brief period, each time producing a minute, passively decaying electronic potential. *b*, As might occur if channels could be "switched on" by drug.

would be of great interest to test whether the amplitude of the elementary event is the same for drugs differing in efficacy, as would be expected of the type of mechanism I have discussed. It should be noted that the assumption about the shape of the elementary event is not the only one possible. The decaying potential considered by Katz and Miledi would correspond to a very brief conductance increase (Fig. 3*a*) and they suggest that each of these might represent the impact of a single drug molecule. The noise could, however, represent the "stepping" of individual receptors from an inactive to an active state, which could result in conductance and potential fluctuations of the type indicated in Fig. 3*b*. Even if the magnitude of the conductance increase associated with each step were constant, the fact that the lifetime of the activated state would not be constant would mean that the amplitude of the depolarization associated with each step would vary. If, as the occupation theory of drug action suggests, activation of a receptor corresponds with the period for which it is occupied by a drug molecule, this latter model would be more appropriate than that used by Katz and Miledi. The allosteric theory of drug action (discussed later) also postulates an activation mechanism more consistent with the type of noise shown in Fig. 3*b* than that of Fig. 3*a*. In this theory, though, the activated state does not coincide with receptor occupation, so the kinetics of the binding of drug molecules could not be determined from analysis of the membrane noise. The evidence so far available does not distinguish between these various theoretical possibilities, but further analysis of the frequency characteristics of the noise should, in principle, enable the distinction to be made.

The view that activation of a receptor represents a quantal rather than a graded transition is implicit in two recent theories of drug action. The first of these was the rate theory³⁹, in which activation was postulated to be a transitory event coinciding with the moment of association between the drug and the receptor. Paton³⁹ suggested that the "quantum" of activation could be the same for all drugs that act on a given receptor, so that the efficacy of a drug would be related to the rate of turnover associated with a given average occupancy and would thus be determined by the dissociation rate constant of the drug-receptor complex. Unfortunately, the experimental evidence is at present inconclusive, and there is little to be added to previous discussions of this controversial question^{6,10,40,41}.

The second type of theory that involves quantal rather than graded transition of the receptor from an inactive to an active state is closely based on the Monod-Wyman-Changeux model for allosteric properties of enzymes^{18,19} and of haemoglobin²⁰. The extension of this mechanism to drug action has been suggested by Changeux *et al.*²¹ and Karlin²². It is postulated that the two conformational states of the receptor, *R* being the active and *T* the inactive state in Karlin's terminology, exist independently of whether or not a drug molecule is bound to the receptor and that drugs act as agonists or antagonists according to their selective affinity for the *R* or *T* conformation respectively. The basic mechanism is thus



K_{AR} and K_{AT} are the microscopic dissociation constants for binding of *A* to *R* and *T*, and $L = T_0/R_0$ the equilibrium constant of the two states in the absence of ligand. By analogy with allosteric enzymes it is suggested that an antagonist could act competitively if it reacted with the same site as the agonist but differed in having a relatively higher affinity for *T* than for *R*, or allosterically if it combined with a second site, again having a higher affinity for *T* than for *R*. According to this view the efficacy of a drug is determined simply by its relative affinity for the two states, agonists having a preferential affinity for receptors in the *R* conformation. Both Changeux *et al.*²¹ and Karlin²² have taken the analogy with allosteric enzymes further and suggested that individual receptors might be grouped as oligomers (Karlin) or as neighbouring units in a lattice-like network (Changeux *et al.*), with the constraint that units in the same oligomer are forced to adopt the same (*R* or *T*) conformation, or that neighbours in the lattice interact in such a way that they tend to adopt the same conformation, the strength of the interaction being assumed to decrease with distance.

There are two important respects in which these allosteric theories differ from the conventional occupation theory of drug action. First, the fraction of receptors in the activated form is not proportional to the fractional occupancy (that is, the binding function differs from the state function). Second, if subunit interactions are postulated, whether among the units of an oligomer or among the neighbours in a lattice, the binding curve deviates from the simple Langmuir equation, and assumes a sigmoid rather than a hyperbolic form. The dissociation curve of oxyhaemoglobin is a very good example.

The evidence in favour of the allosteric theory has centred around the "cooperative" (that is, sigmoid) nature of the concentration-effect curves for depolarizing substances acting on the electroplax cells of the eel, *Electrophorus electricus*. Changeux *et al.*²¹, Karlin²² and Changeux and Podleski⁴² have shown that for agents such as carbachol, decamethonium, phenyltrimethylammonium the curve is sigmoid (Fig. 4), the slope of the Hill plot being 1.5 to 2 compared with 1.0 for a simple hyperbola (Table 1). Similar results were obtained for acetylcholine on the frog motor endplate by Katz and Thesleff⁴³ and Jenkinson³³, and for GABA on crustacean muscle¹⁷ and on insect muscle⁴⁴. The results with GABA refer to conductance change, whereas the other results refer to potential change. In the electroplax, it is important to realize that the relationship between the conductance change (which is presumably the primary effect of the drug) and the resulting depolarization is not very clear^{45,46} so the use of depolarization as a measure of the drug effect is rather unsatisfactory. In the case of frog muscle, the sigmoid concentration-effect curve for depolarizing drugs^{33,43} has been confirmed using a voltage clamp technique (Fig. 4) which gives a direct measure of membrane conductance change.

There are thus two principal lines of evidence that bear on the distinction between the conventional ("occupation") theory of drug action and the allosteric theory. On the one hand, the evidence from reversible antagonists (Fig. 2) suggests that the Langmuir equation is accurately obeyed while, on the other hand, membrane effects tend to show marked cooperativity (Fig. 4). Unfortunately, neither of these pieces of evidence is decisive. Thus, although studies on reversible antagonism seem to favour the conventional model, the equations for competitive antagonism according to the allosteric model²² although not identical to equation (5) are reasonably compatible with these results, the difference between the predictions of the two models being quantitatively very small. Conversely, the finding of cooperativity, which

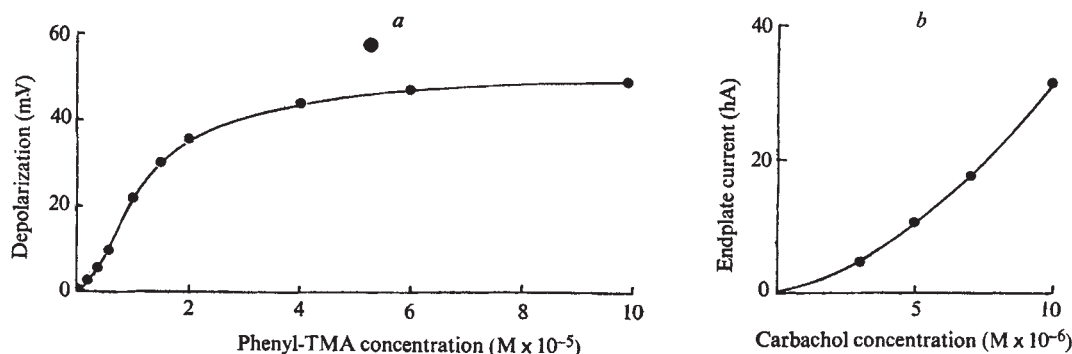


Fig. 4. Results showing sigmoid shape of concentration-effect curves for membrane responses. *a*, Depolarization of eel electroplax by phenyl-trimethylammonium. The concavity at the foot of the curve is characteristic of the action of depolarizing drugs (from ref. 42). *b*, Increase in membrane conductance at voltage-clamped frog motor endplate. Only the foot of the curve is shown because the technique does not permit large changes to be monitored Rang (unpublished).

might seem to favour the allosteric model can be accounted for without difficulty within the conventional framework if an intermediate step is postulated between the primary disturbance of the receptor caused by the bound drug molecule and the consequent membrane permeability change. For example, it might be necessary for one or more out of a group of n receptors to be occupied in order for a given permeability channel to be opened. With such a mechanism the relationship between concentration and conductance change could be markedly sigmoid even though the binding function was accurately represented by the Langmuir equation.

It is interesting that these theories are formally analogous to two postulated mechanisms for allosteric transitions in enzymes, namely Koshland's⁴⁷ "induced fit" mechanism (which corresponds with the conventional occupation theory) and the Monod-Wyman-Changeux¹⁹ "plausible model" (from which the allosteric theory was derived). As has been pointed out⁴⁸, measurements at equilibrium do not unequivocally distinguish between these mechanisms and it seems likely that we shall need (*a*) to measure drug binding directly and (*b*) to follow the kinetics of receptor activation, using the kind of methods developed for studies on enzymes⁴⁹ before the problem of the mechanism of drug action can be resolved.

Isolation of Receptors

More than forty years ago, A. J. Clark⁵⁰ used an indirect method to estimate the binding of acetylcholine in frog heart muscle and concluded that the amount that had to be bound to produce an effect was very small (about 10 pmol/g cells). Since then there have been numerous attempts to detect the binding of drugs to receptors and to isolate the receptor material, so far with only very limited success (reviewed by Lewis and Miller⁵¹). The well known studies of Chagas⁵² and Ehrenpreis⁵³ and their co-workers on the binding of labelled tubocurarine disappointingly revealed only non-specific binding components.

More recently, Paton and I⁵⁴ measured the binding of atropine and related compounds by intestinal smooth muscle and found a saturable component that had the same very high affinity for atropine and other compounds as was found for the receptors (using the dose ratio procedure described earlier). The capacity of this system was 180 pmol/g wet weight or about 2×10^5 molecules per cell. The rate of uptake and washout of atropine was, however, slower than the rate at which the receptor block developed, and it is not clear what was the cause of this discrepancy, though Thron and Waud⁵⁵ have suggested a possible explanation in terms of diffusion-limited kinetics. If this measured uptake does represent binding to receptors as the equilibrium measurements strongly suggest, then this system may be a good one for receptor isolation, as the very high affinity of atropine-like drugs for receptors (equilibrium constants in the range 10^{-8} to 10^{-10} M) should enable the receptors to be labelled relatively specifically.

Potter⁵⁶ described very similar studies using labelled propranolol in an attempt to detect β -receptors in guinea-pig atria, but here the specific uptake seemed to be very small, and was detectable only after subcellular fractionation of the tissue.

Recently, O'Brien and his colleagues^{57,58} have obtained promising results by studying the binding of cholinergic agonists to subcellular fractions of electric tissue from *Torpedo*. They have used equilibrium dialysis to measure the affinity of numerous different drugs for the binding material, and have shown that all of the compounds known to act on cholinergic receptors inhibited the binding of tritiated muscarone (a stable cholinergic agonist), whereas a wide range of other drugs was without any effect. There was quite good correlation between the potency of substances as agonists and their ability to inhibit muscarone binding. These results suggest strongly that O'Brien and his co-workers are indeed on the track of the receptor, though it should be borne in mind that the affinity of agonists for receptors cannot be inferred directly from their potency²⁷⁻²⁹ as the authors assume in interpreting their binding data. They have not yet succeeded in solubilizing the component responsible for drug binding, which is associated with a particulate fraction of the tissue, but the effects of hydrolytic enzymes suggest that the component is a protein or protein-phospholipid complex.

The use of reversible ligands for receptor isolation carries with it the somewhat imponderable danger that the isolation procedures will substantially modify the binding characteristics of the receptor and so interfere with the recognition of it by means of equilibrium dialysis. There are therefore advantages in using irreversible alkylating agents (affinity labels^{59,60}) rather than reversible ligands for receptor-labelling studies, since the receptors can then be labelled in the intact tissue, and the stable radioactive complex studied biochemically. The best known receptor alkylating agents are the catecholamine antagonists related to dibenamine, and several attempts have been made to label receptors with these drugs (reviewed by Lewis and Miller⁵¹). In general, it seems that the specificity of these drugs is low, so that non-specific binding is a serious problem. Various procedures with reversible protecting agents have been devised to improve specificity, but with rather discouraging results^{61,62}. Recently Karlin and his co-workers⁶³ reported preliminary results on the uptake of the quaternary ammonium compound 4-maleimido benzyl trimethylammonium (MBTA) which irreversibly blocks cholinergic receptors of eel electroplax cells. This agent acts only after the dithiol groups have been reduced to free sulphhydryl residues⁶⁴, and some ingenious ways of exploiting this property to improve the specificity of MBTA binding have been devised⁶⁵. So far the uptake of ³H-MBTA only by intact cells has been studied, and biochemical fractionation may well bring about a substantial improvement in the low specificity so far attained.

Other affinity labels have been designed that react very

strongly with cholinergic receptors in the unreduced state. Benzilylcholine mustard (BCM)⁶⁵ is an alkylating derivative of the reversible atropine-like drug, benzilylcholine, and acts at low concentration (10^{-9} – 10^{-8} M) to produce an irreversible block of muscarinic receptors. Preliminary studies on the uptake of ³H-BCM by smooth muscle⁶⁶ gave encouraging results, the uptake being a good deal more specific than that of MBTA by the electroplax, and work on receptor isolation with this system is continuing. A related compound, dinaphthyl-decamethonium mustard^{67,68}, has been investigated recently. This substance acts irreversibly as an antagonist at the motor endplate, and has the interesting property that its affinity for receptors is increased when the receptors are "desensitized" in the presence of a cholinergic agonist^{68,69}. It is hoped that this compound may also prove to be a useful affinity label for isolating receptors.

Two recent reports have shown that a protein component, α -bungarotoxin, of the venom of the snake, *Bungarus multicinctus*, may be a useful receptor-labelling agent. This protein has been extensively studied by Lee and his co-workers^{70,71}, who have shown that it has an irreversible curare-like action on the postsynaptic membrane of the neuromuscular junction, and Lee and Tseng⁷¹ showed that the ¹²⁵I-labelled protein was specifically taken up at the junctional region of mouse diaphragm muscle. Recently Changeux, Kasai and Lee⁷² showed that it also blocked the action of acetylcholine-like depolarizing drugs in eel electric tissue and found that it inhibited the binding of decamethonium to a protein extract prepared from the innervated membranes of electroplax cells. Other studies on the binding of decamethonium to this material⁷² showed that it had a number of properties characteristic of acetylcholine receptors.

Following this lead, Miledi, Molinoff and Potter⁷³ reported the binding of ¹²⁵I-labelled α -bungarotoxin to a protein component of cell membranes from *Torpedo* electric organs, which they solubilized by treatment with a detergent. Analysis by density gradient centrifugation and gel filtration suggested that the binding protein was built of subunits of molecular weight about 80,000. The evidence in favour of identifying this binding material with the receptor protein is at present somewhat incomplete, resting mainly on the finding that tubocurarine and carbachol retard the binding of α -bungarotoxin, whereas physostigmine does not. It appeared that tubocurarine was surprisingly weak as a protecting agent, compared with carbachol, but because there is no information on the effectiveness of these drugs in protecting the receptors against α -bungarotoxin in the intact tissue, the strength of this evidence is not easy to assess. Further studies on this system will be awaited with interest.

Thus, in spite of a number of false starts, there seems every reason to hope that studies in progress will lead to the successful isolation and characterization of cholinergic receptors. This will, I believe, prove to be an important milestone in pharmacology, because we shall for the first time be able to study directly instead of indirectly the chemical interaction between drugs and tissues.

I thank Dr D. Colquhoun for numerous discussions about the topics covered in this article.

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RNA-dependent DNA Polymerase Activity in Virus-like Particles isolated from Human Milk

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Particles apparently similar to type B mouse mammary tumour virus, which are found in human milk and may be associated with breast cancer, contain reverse transcriptase activity. Milks lacking the particles do not have this activity. This finding will help to establish the relationship between these particles and tumour viruses.

PARTICLES morphologically identical to the type B mouse mammary tumour virus have been observed in human milk and in human mammary carcinomas¹⁻⁶. In a recent epidemiological survey² such particles were readily detected in the milk of 60% of American women with a familial history of breast cancer. In American women with no such familial history, particles were found with a frequency of only 5% and when detected were usually present in much lower amounts. They were also found in considerable quantities in 39% of Parsi women of Bombay, a group that has a two to three times higher incidence⁷ of breast cancer than the rest of the Bombay population. Breast cancer accounts for about half of all cancer in Parsi women⁸. The Parsis are descendants of Zoroastrians who emigrated from Persia some 1,300 yr ago and who, because of their strict religious rules, have been inbreeding intensively ever since.

Some of the milks contained particles in quantities sufficient to permit isolation and examination of their enzyme content. We report now a survey of thirteen human milks: all milks positive for type B particles by electron microscopy contain particles that band at densities identical to those of the known oncogenic RNA viruses (1.16–1.19 g/ml.) and exhibit the RNA-dependent DNA polymerase, or “reverse transcriptase”, characteristic of these tumour viruses. All milks negative for particles were also devoid of detectable reverse transcriptase.

Detection of RNA-dependent DNA Polymerase

Unfortunately, a considerable amount of confusion has been generated by the use of assays for this enzyme which have ignored its operational definition. The RNA-dependent DNA polymerase can mediate the synthesis of DNA on a single stranded RNA template. It was initially detected⁹⁻¹¹ in oncogenic RNA viruses by a DNA synthesis that is obliterated

by RNAase and which requires the presence of all four deoxyribonucleoside triphosphates. It was subsequently shown¹² that synthetic duplexes, such as rA·rU; dT·rA; dC·dG, are superior templates for reverse transcriptase.

These synthetic templates serve as useful auxiliary devices in following enzyme activity. But response to them does not constitute unambiguous evidence that a reverse transcriptase has been identified. Indeed, it has been known for some time that the DNA polymerase of *Escherichia coli* accepts rA·rU as a template for the synthesis of dA·dT¹³. Furthermore, DNA polymerases have been found in both cancer and normal embryonic cells which cannot accept pure single stranded RNA as a template but do respond excellently to one or another of the synthetic duplexes (S. Spiegelman, manuscript in preparation). Claims¹⁴ of the universal presence of reverse transcriptase that are based solely on the use of synthetic templates cannot be accepted as definitive.

To avoid this ambiguity, we used two criteria to search for reverse transcriptase in suspected milks, that is, sensitivity of an endogenous reaction to RNAase and a requirement for all four deoxyribonucleoside triphosphates. The synthetic template dT·rA was used only to provide supplementary information.

Purification of Type B Particles

Milk was purified by a modification of the method of Moore *et al.*¹, and stored and purified at 4° C. Some contained the antibiotic ‘Gentamicin’ (Schering) (0.1 mg/ml. milk) as a preservative. Samples were never frozen because this greatly reduces the RNA-instructed DNA polymerase activity (our unpublished observations) and the infectivity¹⁵ of the mouse mammary tumour virus. An equal volume of 0.15 M EDTA (pH 7.5) was added and the milk was centrifuged at 3,000g for 20 min. The clear zone between the lipid and precipitated casein layers was removed and placed on a 20% glycerol column resting on a 100% glycerol cushion. After centrifugation at 90,000g for 1 h, the material on the glycerol cushion was removed and placed on a 10–60% preformed sucrose gradient prepared with TNE buffer (0.01 M Tris, pH 8.3, 0.15 M NaCl, 0.002 M EDTA). After centrifugation at 90,000g for 16 h, fractions were collected and the absorbance at 260 nm determined. The fractions to be assayed for polymerase activity were pooled, centrifuged at 90,000g for 30 min, and resuspended in 0.15 ml. of TNE. This procedure usually resulted in a 500-fold concentration of the starting material.

Many human milks contain structures that resemble mycoplasma² and these were encountered in the present survey. Numerous attempts to propagate these structures have been unsuccessful (unpublished observations of J. Tumilowicz and D. H. Moore). We have examined representatives of this

group for DNA polymerase activities. Six pure cultures of mycoplasmas, four of human and two of animal origin, were examined at protein concentrations and in conditions equivalent to that used in the milk assays. Four showed detectable endogenous DNA polymerase activities (between 0.05 and 0.28 pmol of ^3H -TMP incorporation in 60 min per 100 μg of protein) which were not inhibited by RNAase treatment. All but one showed a significant response to dT·rA, ranging from 0.13 to 6.5 pmol of ^3H -TMP incorporated per 100 μg of protein in 60 min.

These tests were carried out with purified mycoplasma at concentrations exceeding those which would be encountered as milk contaminants by several orders of magnitude. This, and the absence of the diagnostic inhibition of the endogenous reaction by RNAase, gave assurance that the presence of mycoplasma would not interfere with the interpretation of the results. (But sole dependence on the response to dT·rA could introduce an element of ambiguity with heavily contaminated samples.)

Centrifugation Profiles of Clarified Milks

Figs. 1 and 2 show the profiles obtained in sucrose equilibrium gradients of clarified milks that were positive for type B particles by electron microscopy. Fig. 1 is derived from an American woman whose mother and maternal aunt had breast cancer. There is a peak at a density of 1.175 g/ml. with a shoulder between 1.18 and 1.19 g/ml. and another small peak at a density of 1.24 g/ml. Fig. 2 results from a similar centrifugation of a Parsi milk; this shows a principal peak at 1.19, another at 1.16, and a minor component at 1.11. The extensive amount of material absorbing at 260 nm indicates that the particles possessing the RNA-instructed DNA polymerase are still in the presence of contaminating debris.

Milks that are negative for type B particles by electron microscopy often show bands at densities lying between 1.24 and 1.21 g/ml. and also between 1.11 and 1.17 g/ml. None showed distinct peaks near 1.19, the density characteristic of type B particles and the region which shows the highest level of RNA-dependent DNA polymerase activity when it is present. The bands lying outside the 1.16 to 1.19 density region were often positive for endogenous or dT·rA stimulated DNA polymerase, but negative for RNAase sensitivity. Only the

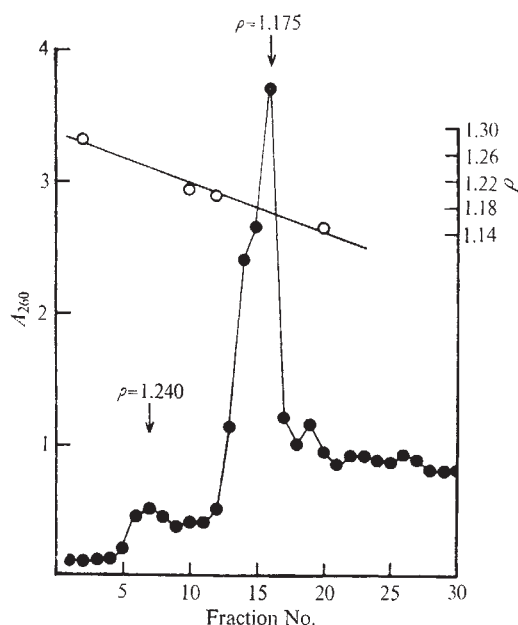


Fig. 1 Sucrose equilibrium gradient centrifugation of human milk particles. 125 ml. of milk positive for type B particles was purified as described in text. This woman's mother and maternal aunt had breast cancer.

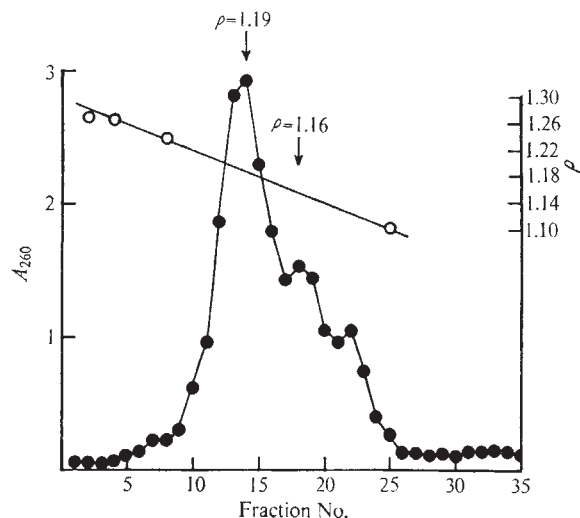


Fig. 2 Sucrose equilibrium gradient centrifugation of particles from a Parsi milk. Parsi milk (60 ml.) was purified as described in the text.

bands within the density region of 1.16 to 1.19 g/ml. from milks positive for type B particles by electron microscopy exhibited an endogenous DNA polymerase activity that was eliminated by the addition of ribonuclease.

Reverse Transcriptase Activity

As is the case with oncogenic RNA tumour viruses, we found that RNAase-sensitive endogenous DNA polymerase activity requires pretreatment with a non-ionic detergent ('Nonidet P-40'). Figs. 3a and b show the kinetics typical of the DNA polymerase activity found in particles isolated from the 1.17 to 1.19 region of the density gradient (Fig. 1). There is an endogenous reaction that continues for at least 1 h and this is virtually eliminated by omission of one of the deoxyribonucleoside triphosphates or the inclusion of ribonuclease in the reaction mixture. These particles also contain a DNA polymerase activity that responds to the synthetic template dT·rA.

Table 1 RNA-dependent DNA Polymerase Activity in Virus-like Particles isolated from Human Milk

Sample No.	Particles	RNA-dependent DNA polymerase
1-350	—	—
2-362A	+	+
3-364	—	—
4-367	—	—
5-368A	—	—
6-368B	+	+
7-369A	—	—
8-369B	—	—
9-369C	+	+
10-370	—	—
11-373	+	+
12-374A	—	—
13-374B	—	—

Milks were processed and purified bands in sucrose equilibrium density gradients were assayed for RNA-dependent DNA polymerase as described. Milks were examined by electron microscopy for the presence of particles as previously described¹.

The particles of the Parsi milk respond in very much the same way to omission of any one of the deoxyribonucleoside triphosphates and to the presence of ribonuclease. Fig. 4 shows that the 1.19 g/ml. and 1.16 g/ml. regions of Fig. 2 contain polymerase activity. Both dT·rA templated and endogenous polymerase activities were present in the 1.19 region at approx-

imately three times the levels (per μg of protein) of those in the 1.16 g/ml. region.

Thirteen milks were examined for type B particles by electron microscopy and corresponding RNA-dependent DNA polymerase assays performed on the material isolated from the 1.16 to 1.19 g/ml. density region of the gradient. Table 1 summarizes the outcome of this comparison and shows that the correlation is complete. Those milks that are devoid of type B particles exhibit no sign of an endogenous DNA-synthesizing ability sensitive to ribonuclease and requiring the presence of all four deoxyribonucleoside triphosphates. On the other hand, the reverse transcriptase was found in the proper density region

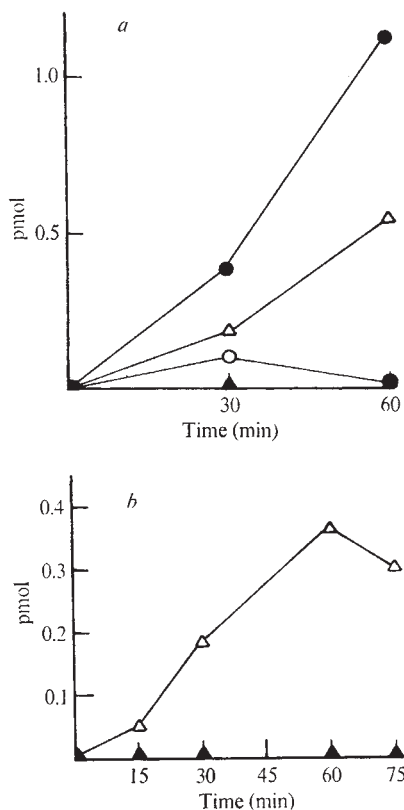


Fig. 3 Characterization of RNA-dependent DNA polymerase activity of human milk particles. *a* illustrates the kinetics of incorporation of ^3H -dTTP into acid-insoluble product employing the purified particles isolated from the milk of a woman with a familial history of breast cancer. *b* illustrates these kinetics using an aliquot of the same purified sample kept at 4°C for 48 h. The standard 125 μl . endogenous reaction mixture contained in μmol : 8 of Tris HCl (pH 8.3), 1 of MgCl_2 , 6 of KCl, 3 of dithiothreitol, 0.2 each of the unlabelled deoxyribonucleoside triphosphates (dATP, dGTP, dCTP), and 0.01 of dTTP. 25 μl . of ^3H -dTTP (2 mCi/ml.) was added to give a final specific activity of 2,000 c.p.m./pmol. The reaction contained 250 μg protein from concentrated pellets resuspended in TNE and 0.08% 'Nonidet P-40'. The DNA polymerase reaction mixture utilizing dT-rA as a template contained the same concentrations of Tris HCl, MgCl_2 , KCl, dithiothreitol, ^3H -dTTP, 'Nonidet P-40', and protein as the endogenous reaction mixture. It also contained 8 $\mu\text{g}/\text{ml}$. dT-rA, 0.04 μmol of dATP and dTTP, giving a final specific activity of 513 c.p.m./pmol. All reactions were carried out at 37°C . At time of sampling, aliquots were withdrawn and precipitated with 1 ml. of trichloroacetic acid-saturated sodium orthophosphate-saturated sodium pyrophosphate 1:1:1 in the presence of 60 μg of *E. coli* RNA as carrier. After 10 min the precipitable radioactivity was collected on a nitrocellulose filter and dried, and the radioactivity was determined in a liquid scintillation counter using BBOT scintillation fluid. The pmol incorporation of ^3H -TMP into acid-insoluble product per 100 μg of protein is recorded at the times indicated. The ribonuclease A (Worthington) used in all experiments was tested for the presence of contaminating DNAase activity, and was found negative. $\bullet$, +dT-rA; Δ , endogenous R_+ ; $\circ$, -dATP; $\blacktriangle$, +RNAase.

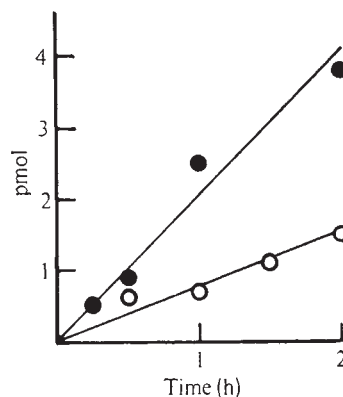


Fig. 4 DNA polymerase activity of purified milk particles. The 1.19 g/ml. ($\bullet$) and 1.16 g/ml. ($\circ$) regions of a sucrose equilibrium density centrifugation were assayed for DNA polymerase activity utilizing dT-rA as template. The results indicate the pmol of incorporation of ^3H -dTTP into acid-insoluble product per 100 μg of protein. The reaction mixture is as described in the legend to Fig. 3.

(1.16 to 1.19 g/ml.) in all milks that were positive for type B particles by electron microscopy.

Human Milk Particle Viruses and Cancer

The human milk particle is indistinguishable in size and ultrastructure from the mouse mammary tumour virus type B particle. It is also similar in many respects to the virus isolated from a spontaneous mammary carcinoma of a rhesus monkey¹⁶ (J. Schlom and S. Spiegelman; R. C. Nowinski, E. Edynak and N. H. Sarkar; B. Kramarski, N. H. Sarkar and D. H. Moore, manuscripts in preparation) and to the particles observed in human breast carcinoma⁶. Particles resembling the oncogenic RNA "type C" particles were occasionally observed in lower amounts in milks positive for the type B particles.

Although the possession of an RNA-dependent DNA polymerase in purified particles (1.16–1.19 g/ml.) is not proof of oncogenic potential, this enzyme activity has been observed in all of twenty-seven isolates of oncogenic RNA viruses¹⁷. Even visna, a virus that causes a "slow" neurological disease in sheep, and the related progressive pneumonia virus of sheep, both of which contain the RNA-dependent DNA polymerase, transform mouse cells *in vitro*¹⁸.

The relationship of the human milk particle to known oncogenic RNA viruses and to human neoplasia is of obvious importance. It will now be possible to resolve by molecular hybridization the relationship between the human type B particles and the similar agents associated with mammary tumours in mice and monkeys. Hybridization of the DNA synthesized by the human type B particle with the RNA isolated from malignant and benign human breast tumours, as well as to "normal" breast tissue, should also answer many questions concerning the relevance of this particle to human breast cancer.

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Post-blastocyst Differentiation *in vitro*

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A technique has been developed for growing mouse embryos *in vitro* to a stage at which post-blastocyst differentiation, including the beginnings of heart beat, can be observed.

HUMAN oocytes can be fertilized and grown *in vitro* to the blastocyst stage, albeit with relatively little success^{1,2}, but no mammalian embryo has been cultured *in vitro* from the blastocyst to the early somite stage. Study of the regulation of development during this critical period has therefore not been feasible. Although it has been established that both oestrogen and progesterone are essential for implantation of mammalian embryos³, the role of uterine tissue in implantation and subsequent development is not well known. The question has arisen whether it is possible to culture mammalian embryos outside the uterus in tissue culture from the pre-implantation stage to the early somite stage, or if uterine tissue is indispensable for the differentiation of mammalian embryos during these periods⁴. The mouse embryo provides a promising model in which to study the post-implantation stage of mammalian embryos *in vitro*, because of the relative ease of obtaining large numbers of eggs at low cost.

Using reconstituted rat tail collagen⁵ as the substrate for implantation, I attempted to culture mouse embryos in a cell-free system.

Pre-implantation

Mouse embryos were collected and prepared according to established procedures⁶. Random bred female mice were made to superovulate by treatment with gonadotrophin; each mouse received 5 IU of pregnant mare serum gonadotrophin ('Equinex', Ayerst), followed 48 h later by 5 IU of human chorionic gonadotrophin (HCG). The mice were caged overnight with males of proven fertility. Insemination was verified the following morning by the finding of a copulatory plug in the vagina. Morula and blastocysts were collected by flushing the uterine horns of the inseminated females with culture medium approxi-

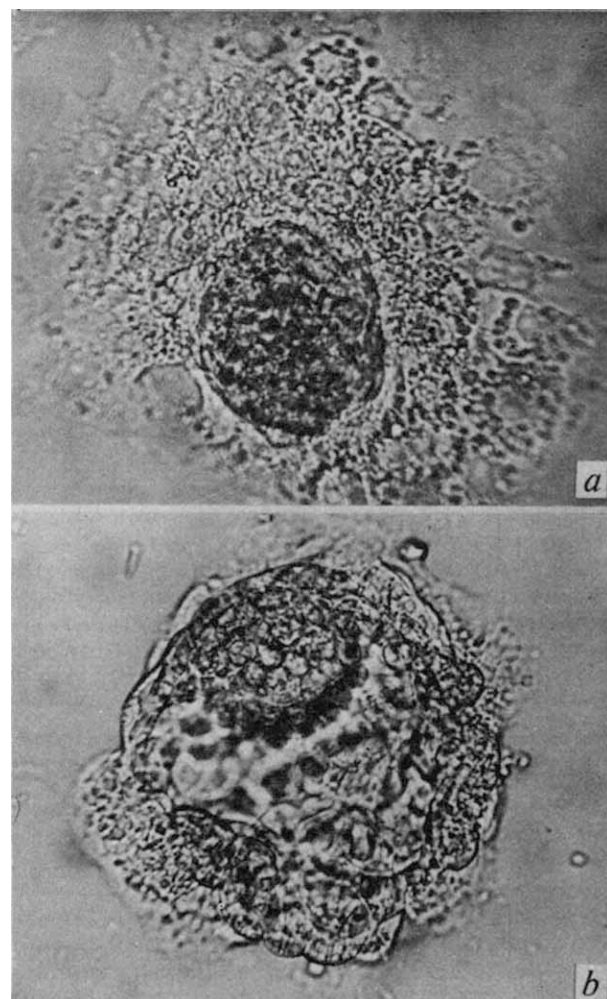


Fig. 1 *a*, Implantation of blastocyst on collagen. Mouse blastocyst after 3 days in culture. Blastocysts which escaped from zona pellucida spread large flat primitive trophoblastic cells into the collagen substrate. The rounded mass in the centre shows the embryo protruding from the surface of the collagen substrate ($\times 280$). *b*, Blastocyst after 4 days in culture, showing formation of trophoblastic giant cells at the periphery and the differentiation of the inner cell mass into ectoderm and proximal endoderm ($\times 280$).

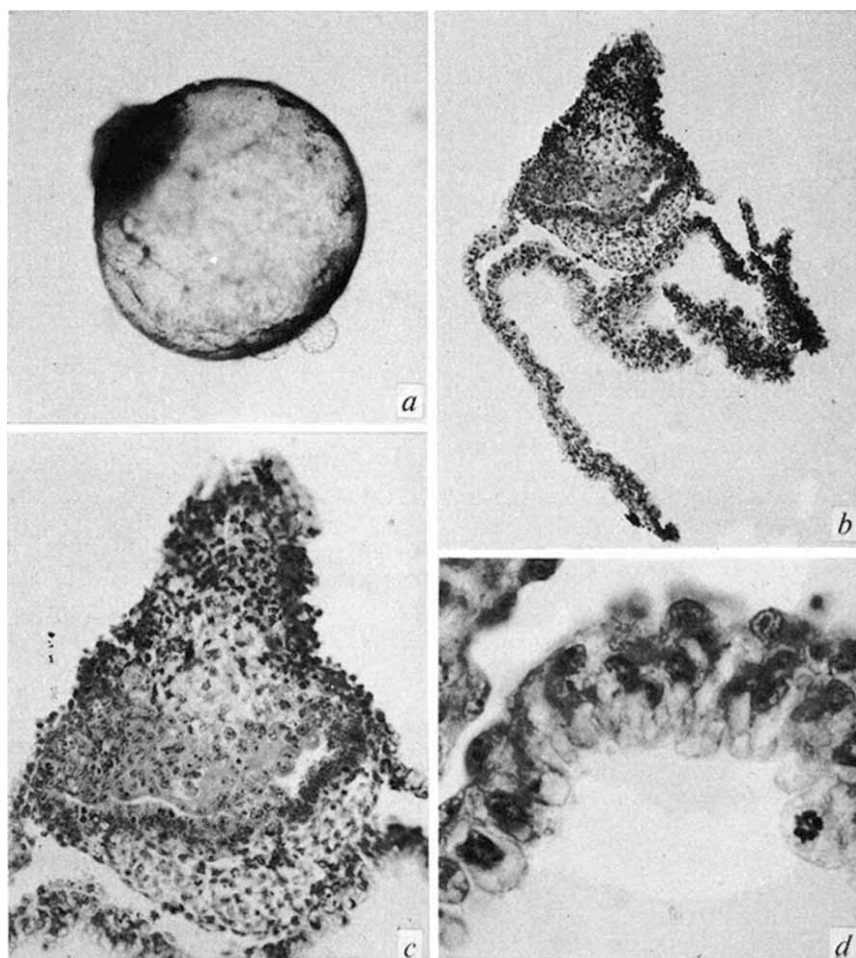
mately 81 h after injection of HCG. Flushings from several mice were collected and pooled in an embryological watchglass after which the flushed embryos were washed and transferred to fresh medium. Groups of thirty to fifty embryos were placed in 50 mm plastic Petri dishes coated with reconstituted rat tail collagen. The culture medium consisted of Eagle's minimum essential medium supplemented with 10% foetal calf serum. The mouse embryos were incubated in a humidified atmosphere of 5% CO₂ and 95% air at 37° C. Many blastocysts began to escape from the zona pellucida on the first day of incubation. The empty zonae, each with its hiatus from which a blastocyst had escaped, remained intact in the medium for several days.

cleavage continued and resulted in an increased number of smaller and smaller cells. After attachment of the embryo, individual cells of the trophoblastic cells increased in size and formed typical trophoblastic giant cells⁷.

Giant-cell Transformation

Some implanted embryos transformed into giant cells, commencing at the abembryonal pole and spreading to the embryonic pole between the fourth and sixth days of incubation (Fig. 1*b*). The inner cell mass developed into an egg cylinder covered with a unicellular layer of endoderm. The proximal

Fig. 2 *a*, Mouse blastocyst cultured for 10 days. After implantation of blastocyst as shown in Fig. 1, the rounded cell mass in centre developed into a large sphere with a diameter of 2–3 mm. The spherical portion floats in the medium, with an ectoplacental cone (upper left) attached to the collagen. Egg cylinder can also be seen in upper left. Vesicles protrude from the surface of the embryo (lower right) ($\times 18$). *b–d*, Histological section of mouse blastocyst after culture for 10 days. Embryo was fixed in 4% formaldehyde; stained with haematoxylin and eosin. *b*, Histological section of mouse embryo of *a*. The yolk sac (lower part) was collapsed during the preparation of this section. Upper part contains the ectoplacental cone and embryonic disk ($\times 90$). *c*, Ectoplacental cone and egg cylinder of *b*. Mitotic figure may be seen at lower centre ($\times 180$). *d*, Enlarged from *b*. Yolk sac consisted of two layers: outer trophoblast and inner endoderm. Active mitotic figure in the trophoblast may be seen at lower right ($\times 750$).



Attachment

On the third day of incubation, each naked blastocyst which had erupted from the zona became attached by its equatorial pole to the collagen and trophoblastic cells began to spread into the underlying collagen. These large, flat cells with vesicular cytoplasm migrated rapidly. Within a few hours, a flat sheet of trophoblastic cells was formed, the embryo remaining visible as a rounded, dense cell mass in the centre of the outgrowth of trophoblast (Fig. 1*a*). It is not known whether this spread of the primitive trophoblastic cells into the substrate has significance in the orientation of inner cell mass and induction of the germinal layer formation immediately following this stage, or whether these cells are the byproduct of embryo damage caused by attachment. Morphologically, these cells were very different from those of the trophoblast observed after the stage of giant cell transformation. Until this stage, the total embryonic mass did not increase significantly, although

endoderm gradually spread to form distal endoderm which underlies trophoblastic giant cells. Between 80 and 95% of the implanted embryos ceased to differentiate at this stage and either grew as a disorganized cell mass or degenerated, a process reported by other laboratories⁸. Only 5–20% of implanted embryos developed beyond the yolk sac stage.

Yolk Sac Formation

The surviving embryos which retained their organization expanded rapidly by the eighth day of incubation and developed into a sphere 2–3 mm in diameter with a protruding ectoplacental cone attached to the underlying collagen (Fig. 2*a*). The spherical portion of the embryo was floating in the culture medium. Fig. 2*b* shows a histological section of a collapsed whole embryo of Fig. 2*a*. The portion of ectoplacental cone and egg cylinder is shown enlarged in Fig. 2*c*. More than three

kinds of cell layers can be differentiated in the egg cylinder: (1) a small undifferentiated extra-embryonic ectoderm; (2) a large differentiated ectoderm, and (3) the proximal endodermal cells. High mitotic activity can often be seen in this area. The yolk sac consists of an outgrowth of the embryonic endoderm from the egg cylinder and lies within the trophoctoderm. At this stage the latter had a relatively large proportion of vesicular cytoplasm and high mitotic activity (Fig. 2d).

Blood Islands, Vessels and Heart Beat

After 10–14 days of cultivation, blood islands developed sporadically on the yolk sac; these contained relatively large primitive red blood cells, distinguishable in the living culture by their brilliant red colour and large size. These blood islands were gradually developed by what appeared to be vascular endothelial cells resulting in the formation of a network. The red colour of the blood cells faded 2–4 days thereafter. Fig. 3 shows red blood cells and blood vessels in developed mesoderm. Rhythmical contractions of 70–80 beats per minute which resembled heart beat were observed in seven of twenty-five post-implantation embryos between 10 and 14 days of cultivation. Red blood cells in blood vessels leading to the contractile area were pumped back and forth. Contractions usually persisted for 2–4 days, then ceased. Vesicular contractions and the movement of red blood cells were recorded with a 16 mm colour movie camera⁹.

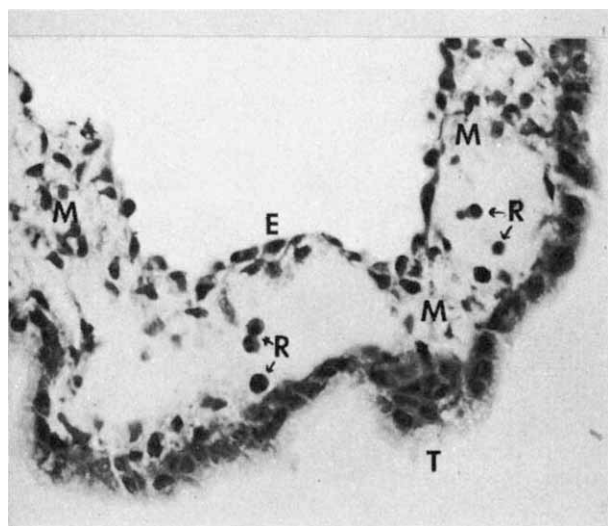


Fig. 3 Blood vessels and primitive red blood cells in mesoderm which developed between trophoctoderm and single cell layer of distal endoderm. (Compare with Fig. 2d.) A large proportion of the red blood cells were lost during preparation of the section. T, Trophoctoderm; E, endoderm; R, red blood cells; M, mesoderm ($\times 350$).

Although implantation and organized post-implantation development has been achieved in organ culture¹⁰, mammalian embryos have been grown in tissue culture during only two phases of development: from the cleavage to the blastocyst stage in mice¹¹ and rabbits¹², and from the early somite to the early limb bud stage in rats and mice¹³. In this report, we have described a degree of success in growing mammalian embryos in tissue culture over the gap between these two periods: from the blastocyst stage to the early somite stage.

The development of each tissue differentiated *in vitro*, however, did not parallel that of *in vivo* development. All embryos cultured *in vitro* by this method seem to be defective in one organ or another. The yolk sac was well developed in one embryo in which blood islands, blood vessels and mesoderm

were formed, yet the egg cylinder was less differentiated than in other embryos. Some embryos developed a vesicular contractility while others formed blood vessels without contractility. The contractions invariably ceased after 3–4 days. The red colour of primitive red blood cells in some embryos faded after 2–4 days of incubation. These changes may be due to the accumulation of the metabolites or to nutritional deficiencies. The nutritional requirement for the outgrowth of mouse blastocysts *in vitro* has been studied. It was found that in addition to glucose and salts, several amino-acids and a macromolecular component were indispensable¹⁴. By modifying the concentration of iron and bivalent cations in the medium, the development of blood vessels and heart beat may be improved. It seems probable that with the present method nutrients are distributed to each tissue of this relatively developed embryo chiefly by the diffusion of nutrients from the culture medium into the embryos, and very little through the primitive vasculature. The supply of nutrients and gases may therefore be inadequate for further embryonic development. It is possible that differentiation may be improved if physical and chemical factors are modified in ways which more closely meet physiological requirements *in vivo*. An increased concentration of oxygen has been used for the cultivation of mouse, rat¹³ and rabbit embryos¹⁰, and the extent of development has been found to depend on the availability of adequate oxygen¹⁵.

Lens fibre from the bovine eye has been used as a substrate for the attachment and implantation of mouse blastocysts *in vitro*¹⁶ and some differentiation has been reported, though little endoderm and no mesoderm were formed. It is not known whether any differences in species, age and physiological status of the animals from which the substrate for implantation was extracted influence the differentiation of culture embryos. Foetal calf serum has been used throughout our experiments, so we cannot exclude the possibility that some serum factor(s), influenced directly or indirectly by hormones, have a role in the implantation and further differentiation of embryos *in vitro*.

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Synthesis of Superheavy Elements in the r -Process

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Contrary to previous opinion, superheavy elements can be produced in the r -process in explosive stellar events. Calculations are based on estimates of the fissionability of very neutron-rich superheavy nuclei using currently accepted mass-law parameters. Certain consequences for empirical cosmic ray abundance and galactic chronology are discussed.

MUCH interest has been aroused by the prediction that the nucleus $^{298}114$ may be a doubly magic nucleus (see Nilsson *et al.*¹), which could mean an island of stability in the mass region near $A \approx 300$. Half lives have been predicted as long as 10^8 yr, but with uncertainties of several orders of magnitude. Thus these nuclei may be of interest with regard to cosmic rays and galactic nucleochronology.

Experimental support for the existence of superheavy nuclei has come from Marinov *et al.*², who believe they have found element $Z=112$ produced in an accelerator. Many workers are also looking for evidence of superheavy nuclei in cosmic rays³, meteorites⁴, lunar material⁵ and terrestrial ores⁶.

The possibility of the stellar synthesis of the superheavy nuclei is an important question, with several conflicting opinions^{7,8}. Viola⁷ does not believe that superheavy synthesis is possible because in terrestrial thermonuclear reactions with targets of ^{238}U , ^{242}Pu and ^{243}Am , no nuclei heavier than ^{257}Fm are recovered. This argument has been countered by Berlovich⁸ who notes that the yields of ^{257}Fm are less with targets of ^{242}Pu and ^{243}Am than they are with ^{238}U (ref. 9), although the corresponding numbers of captured neutrons are less for Pu and Am. This implies that the neutron-induced fissionability of nuclei near the beta stability line must be limiting the production. In the r -process, the neutron captures take place at a rapid rate far off the beta stability line at which Berlovich points out that fissionability is less.

Neutron Capture in the r -Process

The problem is thus to determine when neutron capture in the r -process reaches a maximum in atomic mass and the nuclei involved are then recycled because of neutron-induced and spontaneous fission. For the r -process to proceed from proton

number Z to $Z+1$ the following conditions must all hold (see refs. 10 and 11).

- (a), $\lambda_\beta(Z, N) > \lambda_{sf}(Z, N)$
- (b), $\lambda_\beta(Z, N) > \lambda_{nf}(Z, N)$
- (c), $\lambda_{n\gamma}(Z+1, N-1) > \lambda_{nf}(Z-1, N-1)$
- (d), $\lambda_{n\gamma}(Z+1, N-1) > \lambda_\beta(Z+1, N-1)$

where λ_β , $\lambda_{n\gamma}$, λ_{sf} , and λ_{nf} are the probabilities per second for β decay, neutron capture, spontaneous fission, and neutron-induced fission, respectively. At a given proton number Z , the relative abundances are determined by $n\gamma \rightleftharpoons \gamma n$ equilibrium.

The r -process fission cutoff¹⁰ is calculated from the estimate of Hoyle and Fowler¹², which is based on the assumption that nuclear fissility depends primarily on the parameter

$$F = \frac{Z^2/A}{[1 - \kappa(I^2/A^2)]} \quad (1)$$

where $I = N - Z$ and κ is the ratio of the surface symmetry coefficient to the surface term in the empirical mass formula. (For the mass law of Myers and Swiatecki¹³, $\kappa = 1.79$.) Fissionability increases exponentially with F . On general grounds, Hoyle and Fowler¹² used $\kappa \approx 2$ and thought that fission would occur for $F \gtrsim 40$, which yielded $A_{\max} \approx 275$ as a cutoff for fission for the r -process¹⁰. In this work, the r -process fission cutoff is investigated in much greater detail than the general considerations used by previous workers^{7,10,12}.

Myers and Swiatecki¹³ have shown that as well as the liquid-drop fissility parameter, fission barriers can also depend strongly on shell effects. Nilsson *et al.*¹, taking shell structure into account, have calculated spontaneous fission half lives for many heavy and superheavy nuclei. Because it would be too costly to perform Nilsson half life calculations for each nucleus in the r -process chain, a semi-empirical formula was developed following a procedure similar to that of Dorn¹⁴ and Swiatecki¹⁵. The following equation was used.

$$\log_{10}[\tau_1] = C_1[F + s(N, Z)(C_2 + C_3 F) + C_4 \delta_{OE} + C_5 \delta_{OO} + C_6] \quad (2)$$

where F is the fissility already defined, $\delta_{OE} = 1$ for odd A nuclei, and $= 0$ otherwise, $\delta_{OO} = 1$ for odd-odd nuclei, and $= 0$ otherwise, and $s(N, Z)$ is the Myers and Swiatecki¹³ shell correction term. (τ_1 is in seconds.)

As well as the magic numbers of Myers and Swiatecki which include $N = 184$, we used $Z = 114$ and $N = 228$ (compare ref. 16): other shell correction terms¹⁰ were also tried but did not significantly change the results.

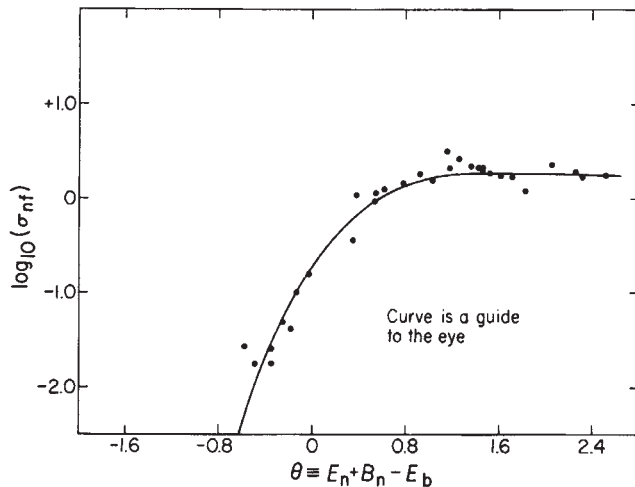


Fig. 1 A plot of $\log_{10}(\sigma_{nf})$ against $\theta = E_n + B_n - E_b$ in MeV, where $\sigma_{nf}(Z, A)$ is the neutron-induced fission cross section in barns, E_n is the neutron energy ($0.1 \text{ MeV} \lesssim E_n \lesssim 1 \text{ MeV}$), B_n is the neutron binding energy in the $(Z, A+1)$ nucleus and E_b is the fission barrier for the $(Z, A+1)$ nucleus. The cross sections are taken from compilations of neutron cross sections^{33,34}. The neutron binding energies are calculated from the nuclear masses, and the fission barriers are either taken from Hyde¹⁸, estimated from cross section data, or estimated from an empirical relationship (equation (3)) using the spontaneous fission half life. Barrier estimates are somewhat arbitrary, thus yielding non-zero values for σ_{nf} when $\theta < 0$. The neutron-induced fission cutoff was therefore chosen as $\theta > -0.8 \text{ MeV}$, rather than $\theta > 0$. This 0.8 MeV can be taken as the difference between the activation energy and the barrier (see ref. 18).

Regression Analysis

The coefficients C_i were found using a least squares multiple regression procedure¹⁷ to fit experimentally known spontaneous fission half lives (Hyde¹⁸; Strutinsky and Pauli¹⁹, excluding the fission isomers; and the measurement of the ²⁵⁸Fm life by Hulet *et al.*²⁰). As well as the experimental points, the theoretical superheavy element half life predictions of Nilsson *et al.*¹ were included in the regression analysis. Terms in F^2 , F^3 and Z^2/A suggested by Dorn¹⁴ were not included because they did not add significantly to the quality of the fit. For consistency, we used the Myers and Swiatecki value for κ of 1.79 which they determined by a fit to the ²⁰¹Tl fission barrier. We found, however, that larger values of κ gave a better fit to the spontaneous fission half lives: fits made with large values predict, for neutron-rich isotopes, shorter spontaneous fission half lives relative to those obtained with small values of κ . To demonstrate the effect of larger values of κ , calculations with $\kappa=2.3$ are also shown. The value $\kappa=2.3$ is representative of typical values used by Seeger²¹. Thus, 1.79–2.3 is a reasonable range for κ .

The best fit coefficients for $\kappa=1.79$ are: $C_1 = -4.34042$; $C_2 = -23.7776$; $C_3 = 0.593943$; $C_4 = -1.28746$; $C_5 = -1.57904$; and $C_6 = -44.6948$; and for $\kappa=2.3$: $C_1 = -4.31583$; $C_2 = -24.0063$; $C_3 = 0.58591$; $C_4 = -1.25604$; $C_5 = -1.663466$; and $C_6 = -45.9078$. This enabled predictions of spontaneous fission lives to within a few orders of magnitude, which is comparable with the accuracy of the Nilsson predictions. Fission barriers can be estimated from the empirical relation (see ref. 13)

$$E_b = 1/8\{29 + \log_{10}[\tau_i(\text{SF})]\} \text{ MeV} \quad (3)$$

with τ in years.

r-Process Fission Cutoff

Neutron-induced fission cross sections $\sigma_{nf}(Z, A)$ for energies in the region of interest to the *r*-process are found to depend on

$\theta \equiv E_n + B_n - E_b$, where E_n is the neutron energy, $0.1 \text{ MeV} \lesssim E_n \lesssim 1 \text{ MeV}$; B_n is the neutron binding energy in the $(Z, A+1)$ nucleus; and E_b is the fission barrier for the $(Z, A+1)$ nucleus (Fig. 1). Theoretically, σ_{nf} (and thus λ_{nf}) should equal 0 when θ is less than 0; however, fission barriers, as well as equation (3), are determined in an arbitrary manner (see caption to Fig. 1). Therefore, we conservatively assumed that σ_{nf} does not vanish until $\theta \lesssim -0.8 \text{ MeV}$.

To obtain the fission cutoffs for the *r*-process a limit was estimated using

$$\log [\tau_i(\text{SF})] \geq \log [\tau_i(\text{SF})_{\text{pred}}] - \log [\Delta\tau_{\text{max}}] \quad (4)$$

where $\tau_i(\text{SF})_{\text{pred}}$ is the value predicted by equation (2) and $\Delta\tau_{\text{max}}$ is the maximum deviation of the predictions of equation (2) from the input data points. The probability is highest that we underestimate A for the fission cutoff. Detailed *r*-process calculations were performed using the basic method of Seeger, Fowler and Clayton¹⁰ with the Myers and Swiatecki¹³ semi-empirical mass law (again with further magic numbers at $Z=114$ and $N=228$). This was rated as the best mass law for *r*-process calculations by Seeger²², giving an excellent fit to *r*-process abundances and having only seven parameters. As well as the mass law, the *r*-process abundance calculation depends on the cycle time (that is, the time to build up to the fissioning nuclei) which is a function of the neutron density, the temperature and the fission cutoff. For a given solution, variations in neutron density and temperature do not change the results as long as the cycle time is constant (see ref. 10). For heavy and superheavy element synthesis, the duration is not important as long as it is much greater than the cycle time.

The cycle time was adjusted by varying the temperature and neutron density so that the calculated *r*-process abundance fitted the observed *r*-process peak at Pt ($A \approx 195$), which produced a cycle time $\approx 8 \text{ s}$. For neutron density $\rho_n = 10^{28} \text{ cm}^{-3}$, this cycle time would correspond to a temperature of $1.8 \times 10^9 \text{ K}$. Each step in the process was tested for the previously outlined conditions (a), (b), (c) and (d). Using the estimated¹⁰ limit for $\tau_i(\text{SF})$ and θ it was found that the *r*-process recycled as a result of fission at $Z_{\text{max}} \approx 104$ and $A_{\text{max}} \approx 307$ (see Fig. 2). Calculations were made with fission lives estimated using a value of κ larger than 1.79. These larger κ calculations yielded fission recycling at slightly smaller values of Z and A . For

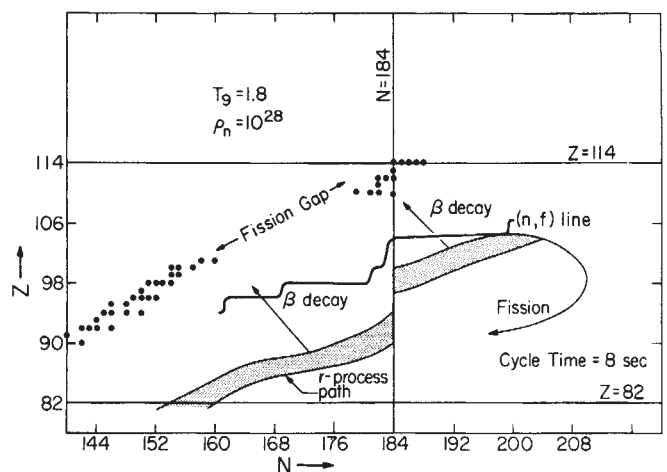


Fig. 2 Path of the *r*-process relative to the long-lived nuclei (the dots). The line labelled (n,f) is the minimum Z for which odd A nuclei have $\theta \geq -0.8 \text{ MeV}$ ($\kappa=1.79$). From this line it is clear that neutron-induced fission causes recycling of the *r*-process at $Z=104$. Although the (n,f) line first intersects the path at $A=301$ and $Z=104$, nuclei with A as large as $A_{\text{max}} \approx 307$ can be synthesized because the (n,f) line does not intersect $Z=103$. The "fission-gap" separating the superheavy "island" of stability from the actinides is shown. An *r*-process abundance peak is produced in this gap by the decay of the nuclei built up in the deviation of the *r*-process flow at $N=184$.

$\kappa=2.3$, $Z_{\max}$ and $A_{\max}$ were reduced to ≈ 102 and ≈ 299 , respectively.

After the termination of the r -process, the neutron-rich superheavy nuclei β decay towards the island of stability near $Z=114$. The superheavy r -process nuclei are made after the jump in the Z value of the r -process path at $N=184$ (Fig. 2). Thus, the number of β decays required to reach β stability is a minimum. In order to reach the "island" it is necessary for $\lambda_{\beta} \gg \lambda_{\text{SF}}$ for each nucleus in the β decay chain. The β decay rates were calculated for these nuclei as in the r -process calculation¹⁰ using $\lambda_{\beta} \propto W_0^6$, where W_0 is the nuclear mass difference. Limits on the spontaneous fission lives were again estimated using equation (4). We found that the r -process products were able to reach the region described by Nilsson *et al.*¹. What happens after reaching the "island" depends on the relative β , α , and SF lives in this region. If the relevant lifetimes were calculated by Nilsson *et al.*¹, we use their estimates: when they were not available, then the lifetimes were calculated as follows: β lifetimes—as discussed here ($\lambda_{\beta} \propto W_0^6$); α lifetimes—from calculated α energy in the manner described by Nilsson *et al.*¹; and SF lifetimes—from equation (2).

Yields of superheavy nuclei with the longest estimated decay lives are shown in Table 1 for both $\kappa=1.79$ and $\kappa=2.3$. Several nuclei with long α and SF lifetimes predicted by Nilsson *et al.* are excluded because of short β lives or shielding by longer lived r -process nuclei. The calculated abundances include sums over α progenitors. (Note that the r -process abundances for the superheavy nuclei are very sensitive to the value of κ .)

It is important to recall that, as stated by Nilsson *et al.*¹, the estimated lives could be inaccurate by several orders of magnitude, but the trends in the half lives should remain valid. These calculations indicate that any superheavy nuclei from the r -process found in cosmic rays should have $110 \leq Z \leq 113$. (Note that the doubly magic nucleus, $^{298}114$, has a predicted α life of only ≈ 10 yr¹, and thus should not be observed.)

Superheavy Nucleus Decay

All superheavy nuclei produced will eventually decay to a spontaneously fissioning nucleus. The delay between production in the r -process and subsequent fission is estimated to be a maximum at $\approx 10^8$ yr for $^{294}110$. This is comparable with the interval between the end of nucleosynthesis and the solidification of objects in the solar system^{23,24}. Long lived superheavy nuclei might thus explain the so called CCF fission spectrum observed in meteorites (compare ref. 25). This spectrum is only observed in the carbonaceous chondrites, which solidified after approximately the same ^{129}I – ^{129}Xe interval as other meteorites not showing the CCF spectrum²⁶. This indicates that the fissioning nucleus (if the effect is a result of fission) was fairly volatile and thus only retained in carbonaceous meteorites. The predicted volatility⁴ for the superheavy nuclei makes

them very likely candidates. The B fission Xe spectrum²⁷ has been definitely identified as coming from extinct ^{244}Pu (E. C. Alexander, R. S. Lewis, J. H. Reynolds, and M. C. Michel, results to be published), and thus a long lived fissioning superheavy nucleus is not necessary for an explanation in this case.

A further interesting by-product from r -process production of super-heavy nuclei is that between the actinides and the superheavy island of stability is a region which we call the "fission gap" ($263 \lesssim A \lesssim 290$). All r -process nuclei made in this region will rapidly β -decay to a spontaneously fissioning nucleus after the r -process has ended. As the r -process has terminated, these fissioning nuclei will not be recycled.

In this fission gap, an r -process abundance peak occurs at $A \approx 281$ (see caption, Fig. 2) which is attributable to the magic number $N=184$ and is similar to the peaks at $A \approx 195$, attributable to $N=126$; $A \approx 130$, attributable to $N=82$; and $A \approx 80$, attributable to $N=50$ (see ref. 28). A reasonable assumption of asymmetric fission could put the fission mass yields from the $A \approx 281$ abundance peak at $A \approx 155$ and $A \approx 126$. The latter would be obscured by the large abundance peak at $A \approx 130$; however, the fission yield at $A \approx 155$ would coincide with a hump in the abundances, previously hard to fit. Although Cameron had proposed²⁹ that fission may explain this feature, most authors^{10,28} had attributed this hump at $A \approx 155$ to nuclear deformations. But quantitative fits to this hump could not be obtained in calculations which fit the other r -process abundance features (see ref. 10). In this work, we are able to fit the standard r -process features and still provide a quantitative explanation of the hump. The sum of the abundances in the hump¹⁰ is ≈ 1.65 ($\text{Si}=10^6$) and the sum of the calculated abundances in the $A \approx 281$ abundance peak is ≈ 1.8 which is consistent with r -process calculations.

Another question which we investigated in these r -process calculations was the values of the r -process production ratios for $^{232}\text{Th}/^{238}\text{U}$, $^{235}\text{U}/^{238}\text{U}$, $^{244}\text{Pu}/^{238}\text{U}$ and $^{237}\text{Np}/^{238}\text{U}$ (Table 2). These ratios are important in nucleochronology calculations^{23,24} and their variations have been discussed by Seeger and Schramm³⁰. The abundance calculated for each progenitor in the decay chains is shown in Table 2: the yields for those progenitors which have a spontaneous fission branch are multiplied by the fraction which α decays. The α -fractions for $A=248, 250, 252$, and 256 are based on experimental results (C. Cowan, unpublished results). Moreover, the α decay fractions for $A=259, 261, 263$ have been estimated by the techniques already used on the superheavy nuclei. Nuclei heavier than $A \approx 263$ (and $A \approx 290$) undergo spontaneous fission rather than decay by α emission. These estimates add progenitors to the odd A chains, ^{235}U and ^{237}Np , relative to the estimates of previous workers³⁰. In view of the uncertainties in the estimates of the α branching ratios, Table 2 also shows the result if nuclei with $A=259, 261, 263$ are assumed to decay completely by spontaneous fission.

Table 1 Long Lived Superheavy Nuclei made in r -Process arranged in Order of Decreasing Half Life

Z	A	$\tau_{1/2}$ (yr)	Dominant decay *	Progenitors †	Abundance ‡ $\kappa=1.79$	($\text{Si}=10^6$) $\kappa=2.3$
110	294	10^8	α	$(294) + 0.13 \times [(298) + (302) + (306)]$	0.030	0.008
113	297	10^5	α	$(297) + (301) + (305)$	0.069	0.010
111	293	10^5	α	$(293) + [(297) + (301) + (305)]$	0.109	0.040
112	295	10^4	α	$(295) + (299) + (303)$	0.087	0.029
112	296	10^4	α	$(296) + (300) + (304)$	0.042	0.002
110	291	10^4	α	$(291) + [(295) + (299) + (303)]$	0.126	0.068
110	292	10^3	SF	$(292) + [(296) + (300) + (304)]$	0.068	0.022
112	294	10^3	α	$0.87 \times [(298) + (302) + (306)]$	0.025	0.000

* All these nuclei eventually decay to spontaneously fissioning nuclei. † a Abundances for $A \geq 300$ (≥ 294 for $\kappa=2.3$) are attenuated by neutron-induced fission during the r -process. (No nuclei are made with $A \geq 307$.) b , The branching for $^{298}112$ is estimated to be 13% α , 87% β^- .
‡ Abundances have been normalized to Pt peak ($A \approx 195$) on $\text{Si}=10^6$ scale. Cycle time for this r -process was 8 s.

Table 2 *r*-Process Yields ($\text{Si}=10^6$); Cycle Time=8 s

232	0.0130	237	0.0209
236	0.0114	241	0.0127
240	0.0167	245	0.0114
244	0.0145	249	0.0147
248×0.917	0.0122	253	0.0114
252×0.892	0.0129	257	0.0064
256×0.070	0.0005	$261 \times (0.71 \text{ or } 0.0)$	0.0078 or 0.0
$\downarrow$		$\downarrow$	
232_{Th}	0.0812	237_{Np}	0.0853 or 0.0775
244	0.0145		
248×0.917	0.0122	235	0.0181
252×0.892	0.0129	239	0.0137
256×0.070	0.0005	243	0.0143
$\downarrow$	0.0401	247	0.0125
244_{Pu}		251	0.0121
		255	0.0111
		$259 \times (0.85 \text{ or } 0.0)$	0.0056 or 0.0
		$263 \times (0.24 \text{ or } 0.0)$	0.0023 or 0.0
238	0.0144	$\downarrow$	
242	0.0129	235_{U}	0.897 or 0.0818
246	0.0169		
250×0.10	0.0013		
$\downarrow$			
238_{U}	0.0455		
$232_{\text{Th}}=1.78$	$244_{\text{Pu}}=0.88$	$235_{\text{U}}=1.97 \text{ or } 1.80$	
238_{U}	238_{U}	$237_{\text{Np}}=1.87 \text{ or } 1.70$	

Fossil Fission Tracks

The validity of the observations of Bhandari *et al.*⁵ is not discussed here; however, it is interesting to use their results as an example of how to apply these *r*-process calculations to experimental observations.

Bhandari *et al.* observed fossil fission tracks from a presumed superheavy source, as well as from ^{244}Pu and ^{238}U . The average track density ratio $[\rho_{\text{tracks}}(\text{superheavy})/\rho_{\text{tracks}}(^{244}\text{Pu})]$ was ≈ 1 for both the lunar fines and meteoritic material, which would yield an implied abundance ratio at the time of track retention of $R \equiv [\text{superheavy}]/[^{244}\text{Pu}] \approx 1.25 \times 10^{-3}$ because each superheavy nucleus would yield fission tracks, and $\tau_a/\tau_{\text{sf}} = 1.25 \times 10^{-3}$ for ^{244}Pu (ref. 31). The implied decay life for the superheavy element can then be calculated assuming an interval, δ , between the end of nucleosynthesis and the time of fission track retention, the value of which is $\approx 5 \times 10^8$ yr as the interval between Xe retention and track retention is $\approx 3.8 \times 10^8$ yr (Wasserburg *et al.*²⁷) and, as mentioned previously, the interval between the termination of nucleosynthesis and Xe retention is $\approx 10^8$ yr.

The *r*-process production abundance, P , for the longest-lived superheavy nucleus $^{294}110$, relative to ^{244}Pu , can be obtained from Tables 1 and 2, $P = 0.030/0.040 = 0.75$ for $\kappa = 1.79$, and $P = 0.2$ for $\kappa = 2.3$, from which the implied decay life for $^{294}110$ can be calculated. For continuous nucleosynthesis (a final sudden event would change the results by less than 10%):

$$R = P \frac{\tau_{\text{SH}}}{\tau_{244}} \exp \left[\frac{0.693\delta}{\tau_{244}} \left(1 - \frac{\tau_{244}}{\tau_{\text{SH}}} \right) \right]$$

which yields, for $\kappa = 1.79$,

$$\tau_{\text{SH}} = \frac{\tau_{244}}{1 - \frac{\tau_{244}}{0.693\delta} \left[\ln(1.67 \times 10^{-3}) + \ln \left(\frac{\tau_{244}}{\tau_{\text{SH}}} \right) \right]} \approx 3.5 \times 10^7 \text{ yr}$$

where τ_{SH} is the superheavy half life and $\tau_{244} = 8.18 \times 10^7$ yr (ref. 31): for $\kappa = 2.3$, $\tau_{\text{SH}} \approx 4 \times 10^7$ yr. These results are within a factor of three of the Nilsson predictions for $^{294}110$ and thus are well within the bounds of uncertainty involved.

A generalization of this result is that all the Nilsson predictions should be reduced by a factor of three. The lifetime of cosmic rays in the galactic disk is $\approx 10^6$ yr (ref. 32). Table 1

therefore indicates that the nucleus $^{294}110$ would be expected to be found in the cosmic rays, but no other superheavy nuclei ($Z \gtrsim 100$) should be observed. This conclusion is subject to qualification, however, because of the uncertainty involved in both our model and the calculation of fission half lives.

The results shown in Tables 1 and 2 can be used to date the cosmic rays. If the ratio [Superheavy nuclei]/[Actinide nuclei] is $\gtrsim 0.2$ in cosmic rays, the age of the heavy cosmic rays is $\lesssim$ the half life of $^{293}111$, $\approx 10^5$ yr.

In conclusion, if the *r*-process fission cutoff is estimated with shell structure taken into account, it is possible to synthesize superheavy elements. This result is sensitive to the value of κ (the surface symmetry coefficient/the surface energy coefficient), and it also explains the abundance hump at $A \approx 155$ as attributable to the fissioning of the $N = 184$ abundance peak produced in the "fission gap".

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LETTERS TO NATURE

PHYSICAL SCIENCES

X-rays from the Coma Cluster of Galaxies

THE Coma cluster of galaxies is usually considered to contain insufficient matter in the form of galaxies to be gravitationally bound, but the high degree of symmetry of the cluster, and the presumed age of its galaxies, suggest that the cluster is, in fact, gravitationally bound. It is possible that the missing mass takes the form of ionized intracluster gas, and Woolf¹ has used radio, visible light and X-ray observations to set upper limits for the amount of such matter and, more recently, Turnrose and Rood² have discussed the problems of heating and supporting such a gas against gravitational collapse. We present here new observational evidence of X-ray emission which limits the amount of hot intracluster gas to less than 2% of that required for gravitational binding.

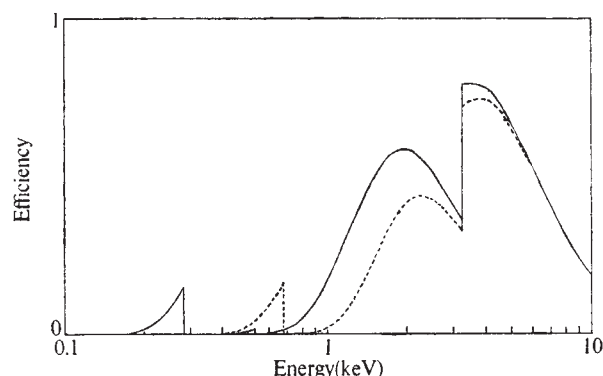


Fig. 1 The efficiency of the X-ray proportional counters as a function of photon energy. The solid curve is for the 'Mylar' window detector and the dashed curve is for the 'Teflon' window detector.

On March 14, 1969, at 0530 UT an Aerobee 150 was launched from White Sands Missile Range, New Mexico. Aboard the rocket were two proportional counters filled to about 800 mm Hg with 10% methane in argon. The windows of the two proportional counters were 'Teflon' (3.2 μm) and 'Mylar' (3.8 μm) with open areas of 234 cm^2 and 326 cm^2 , respectively. The quantum efficiencies of the two detectors are shown in Fig. 1. During the flight, the rocket performed a slow scan, such that the detector field of view moved at a rate of approximately $0.35^\circ \text{ s}^{-1}$ over the Coma cluster and adjacent sky. Each detector had a cylindrically symmetrical field of view of about 10° full width at half maximum and about 20° full width at cutoff.

Table 1 lists the results of three observations each lasting 27 s; one observation was centred approximately on the Coma cluster, and the two adjacent positions gave background data. The data shown are the count totals from the detectors for the approximate bandwidths indicated and are uncorrected for instrumental field of view, area, and quantum efficiency.

Table 1 Count Rates near Coma Cluster

Position	Observed counts per 27 s				Total
	18–24 Å	'Teflon' 1–12 Å	'Mylar' 44–60 Å	1–17 Å	
Before Coma	169	1,821	242	2,732	4,964
Coma	202	1,900	259	3,092	5,453
After Coma	208	1,872	248	2,865	5,193

Fig. 2 shows the result of a pulse height analysis of the data, corrected for field of view and area; the mean of the count totals from the two background intervals has been subtracted from that obtained from the region which includes the Coma cluster. It is clear from Table 1 and Fig. 2 that only upper limits of the flux in the 44–60 Å and 18–24 Å bands from the Coma cluster can be specified, but the flux measured from 3–17 Å is statistically significant. The last column in Table 1 shows the sum of the other columns and indicates an approximately 5σ X-ray enhancement over the sky background from the direction of the Coma cluster.

The 'Teflon' counter taken separately indicates an enhancement of only 1σ , although the 'Mylar' counter shows a 4.5σ increase above background. Almost all this increase occurs in

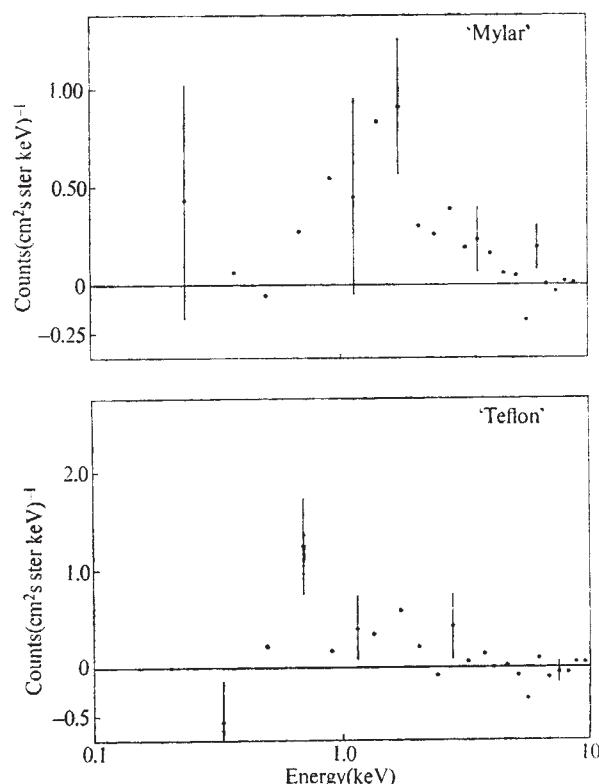


Fig. 2 Spectra from the two detectors of the Coma cluster region after subtraction of the diffuse background. Vertical error bars are 2σ in length and represent the combined uncertainty in the background and Coma cluster data. The solid angle assumed is that of the detector (0.024 ster).

the energy band 0.7–4 keV (centred at 8 Å) in which the efficiency of the 'Mylar' counter is considerably higher than that of the 'Teflon' counter (Figs. 1 and 2). Over the entire 0.7–4 keV bandwidth, the Coma X-ray flux is approximately 0.03 counts cm⁻² s⁻¹, a factor of ten below the upper limit (at the 3σ confidence level) set by Friedman and Byram³ in a similar energy band.

From the data shown in Fig. 2, and because the Coma cluster subtends a solid angle of about 2.5×10^{-3} steradians, upper limits have been placed on the flux in the three bands of 44–60 Å (from the 'Mylar' detector), 18–24 Å (from the 'Teflon' detector) and 1–3 Å (from both detectors). The flux from the 3–17 Å (0.7–4 keV) band is a positive detection. The values are listed in Table 2.

Table 2 Photon Flux from Coma

Wavelength band (Å)	Flux (photons cm ⁻² s ⁻¹ ster ⁻¹ keV ⁻¹)
44–60	450 (upper limit)
18–24	250 (upper limit)
3–17	11 (observed)
1–3	20 (upper limit)

We can use the X-ray data to test the hypothesis that the cluster is gravitationally bound by an optically thin, hot intracluster gas, as was done by Woolf¹. From the observed flux (3–17 Å) and the upper limit in the longest wavelength band (44–60 Å) we obtain a minimum temperature of 8×10^6 K. At this temperature, the mass of gas required to bind the cluster ($\sim 3 \times 10^{48}$ g) would radiate much more intensely than the observations permit. Only if the total mass is less than 6×10^{46} g or about 2% of that required for gravitational binding can the X-ray observations in the various wavelength bands be reconciled. This estimate assumes a uniform distribution of gas. Concentration in clouds would decrease the mass still further. Taking the diameter of the cluster to be about 4°, or 5 Mparsec, gives a density upper limit of about 2×10^{-5} cm⁻³, which is close to the critical density for intergalactic gas in a closed universe.

The possibility that individual galaxies within the Coma cluster are responsible for the observed X-rays has been shown to be unlikely⁴. We will assume that the population of X-ray sources in our galaxy is typical of all galaxies of similar optical luminosity and compare the X-ray emission of our galaxy with that of the Coma and Virgo clusters. (The following gross comparison does not take into account that both the Virgo and Coma clusters include a great number of elliptical galaxies, whereas ours is a spiral galaxy.) The Virgo and Coma clusters contain of the order of 100 and 1,000 galaxies, respectively, at least as bright as our own. The estimated power of our galaxy from discrete X-ray sources is about 7×10^{39} erg s⁻¹ for the 1–10 Å region⁵. At a distance of 70 Mparsec the expected X-ray flux from the 1,000 bright galaxies in the Coma cluster would be about 5×10^{-3} photons cm⁻² s⁻¹ or about a factor of twenty below that observed. Byram, Chubb, and Friedman⁶ found that the X-ray power of the Virgo cluster (minus M-87) is about 1.6×10^{43} erg s⁻¹ in the 1–10 keV range and suggested that it could be accounted for as the integral of all galaxies in a field of view of approximately 30 square degrees. It was an oversimplification, however, to treat each of 2,000 galaxies in Virgo as equivalent to our galaxy in X-ray power. For the approximately 100 bright galaxies in Virgo, the X-ray flux from discrete sources would be only 7×10^{41} erg s⁻¹ (assuming 7×10^{39} erg s⁻¹ galaxy⁻¹), again a factor of twenty below that observed. The X-ray fluxes from both the Virgo and Coma clusters thus seem to be proportional to the optical luminosity of the cluster.

If the estimated X-ray power of our galaxy is integrated over all galaxies within a Hubble radius to obtain their contribution to the diffuse background, the result is a factor ten to fifty times

lower than the observed background. Because the total X-ray flux observed from Coma and Virgo is about an order of magnitude greater than might be expected from discrete sources, every cluster may well contain a mass of hot intracluster gas proportional to the number of bright galaxies in the cluster. The integral over a Hubble radius could then give sufficient X-ray flux in the 0.7–4 keV range to make up most of the diffuse background observed in that portion of the spectrum by Henry, Fritz, Meekins, Chubb, and Friedman⁷. It would still be possible to attribute the flux observed near $\frac{1}{2}$ keV to intergalactic gas at a temperature close to 10^6 K and the critical density of about 10^{-5} cm⁻³.

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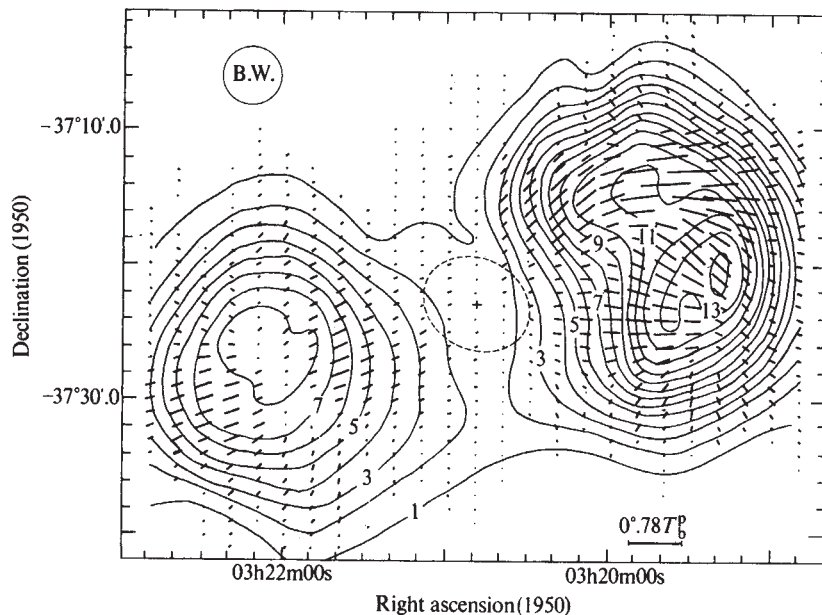
Optical Identification of PKS 0320–37

Lü¹ has suggested that the large extended radio source PKS 0320–37 (Fornax A), identified for many years with the tenth magnitude SO galaxy NGC 1316 (ref. 2), should really be identified with a seventeenth magnitude blue stellar object. We propose to show here why this new identification should not be accepted.

The structure and scale of the radio source are typical of a radio galaxy and not of any known QSO. The total intensity and linear polarization distributions at a wavelength of 6 cm (F. F. G. and J. B. W., work in preparation) are shown in Fig. 1. They were obtained with the Parkes 64 m telescope and a resolution of 4.0'. The distribution consists basically of two highly polarized components (up to 50% linearly polarized) with centres separated by 32'. They straddle the galaxy NGC 1316, which is centred at 03 h 20 m, 47 s, $-37^\circ 23.1'$ (ref. 3). The broken lines represent the outer limits of the optical nucleus as shown on a limiting exposure at visual wavelengths with the 48 inch Palomar Schmidt telescope⁴. The 6 cm radio distribution is similar to that found at 75 cm with similar resolution⁵. The apparent dimensions are typical of a radio galaxy located at the distance of NGC 1316 (about 20 mparsec) but exceeds by two orders of magnitude those of any known QSO.

For the galaxy NGC 1316, evidence of an association of optical and radio continuum has been provided by Arp⁴. Using a faint photography technique, he showed that the galaxy had outer extensions which bend into the two radio components.

Fig. 1 The 6 cm distributions of total intensity and linear polarization over PKS 0320-37 (Fornax A). The directions of the magnetic field in the source are virtually orthogonal to the vectors shown. The contour interval corresponds to a brightness temperature of 0.155 K; the scale of polarization brightness temperature (T_p) is shown on the figure. The outer limits of NGC 1316 are represented by a broken line.



In comparing the positions of radio centroid and optical objects, Lü has misused the position data of the catalogue of Bolton, Gardner, and Mackey⁶. He assumed a position error of 0.6' in each coordinate, the value quoted in the catalogue which is applicable to 90% of the listed sources. But, because of the large angular size of PKS 0320-37, the position of its emission centroid was rounded off to 1', compared with 0.1' for the catalogue sources in general, and the quoted error is thus not applicable. The difference between the centroid position and the galaxy is therefore considered insignificant. Lü's argument based on coincidence of position is also invalid; the parent galaxy is frequently displaced from the centroid of the associated radio emission.

In conclusion, we strongly recommend that the identification of the radio source with NGC 1316 be retained.

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steady state or most other current theories. In particular, a big bang hierarchical cosmological model has been described by Wertz^{3,4}. Possible ways to introduce hierarchical fragmentation in the early stages of such a model have already been suggested by Ozernoy⁵ and others. Alternatively, the hierarchy might develop at a later stage, as proposed by Haggerty⁶.

We hope that cosmologists will not continue to insist that the universe must be homogeneous and isotropic simply because theories are made easier by this assumption. The observational evidence shows that the real universe is not built that way. The sooner this fact is recognized the earlier the "grin cosmologies"⁷ and "toy universes"¹ can be retired to their well-deserved place in the History of Science, next to epicycles.

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Hierarchical Big Bang Cosmology

MCCREA¹ has correctly stated that one of us (G. de V.) had criticized² "current big bang cosmology" in reviewing the overwhelming evidence for hierarchical clustering in the galaxy distribution; it is incorrect, however, to assume that such criticism was directed only or even chiefly at big bang models. The principal intention was to point out the need to study the impact on cosmological theories of hierarchical structure in the distribution of matter. Hierarchical cosmology attempts to replace the idealized homogeneous isotropic models by more realistic ones, which have explicit structure on various large scales. It is in no way opposed, in principle, to big bang,

Super-rotation of Upper Atmosphere

RECENT publications^{1,2} report that the mean rotational speed of the Earth's upper atmosphere at altitudes of about 200-400 km is faster than that of the Earth. Rishbeth³ has suggested that an induced electric polarization field at low latitudes plays a part in this "super-rotation". We offer here an alternative explanation, based on the Scott effect⁴.

This effect can be demonstrated with apparatus such as that outlined in Fig. 1. A gas is contained at a low pressure of the order of 10^{-2} torr in a cylindrical vessel, in which a small cylinder is suspended by a torsion fibre. If the cylinder and vessel wall are maintained at different temperatures and a weak

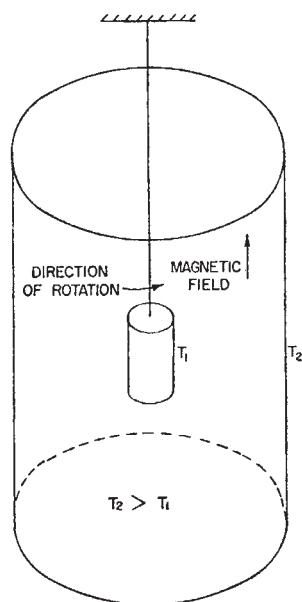


Fig. 1 Schematic diagram of apparatus for laboratory observation of the Scott effect. The relationship between the directions of temperature gradient, magnetic field and gas rotation is shown for molecular nitrogen and similar gases.

axial magnetic field of the order of 1 gauss is applied, there is a resultant torque on the suspended cylinder.

The magnitude and direction of this torque depend on the temperature gradient, the magnetic field and the type of gas. If either the magnetic field or the temperature gradient is reversed, the torque reverses also. This effect is exhibited by several diatomic gases including nitrogen, nitric oxide and several gases present in the atmosphere. With the exception of methane and hydrogen⁵, these all produce a torque in the same sense.

We have subsequently shown that the gas used in these conditions also undergoes bulk rotation in the same sense as the torque on the cylinder, from which we concluded that the Scott effect could contribute angular momentum to astronomical bodies in suitable exterior conditions⁶.

In particular, the upper atmosphere of the Earth—above 100 km—may be such a situation⁷. Here there exist a population of gases, a temperature gradient (temperature increases with height), a low field, of the order of 1 gauss and a low pressure of the order of 10^{-4} torr and less. These parameters conform to the conditions in the laboratory, except that the pressure is two orders of magnitude smaller and the scale is, of course, much larger. Nevertheless, although no thermomagnetic torque measurements have been made at 10^{-4} torr, they do indicate that the direction of the torque remains unchanged as the pressure is reduced below 10^{-2} torr. Furthermore, it must be remembered that the Scott experiments deal with the motion of a sensing body rather than directly with the motion of the gas. Therefore, given the conditions existing in the upper atmosphere, we would expect the Scott effect to produce a west to east rotation, with maximum speed during daylight when the temperature gradient is strongest. This is consistent with the observation that the wind is usually eastward with a speed that is high in the evening and low in the morning¹.

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Nuclear Relaxation Studies of Lecithin Bilayers

BILAYER phases of lipids have broad proton magnetic resonance (PMR) spectra, which have been attributed to incomplete averaging of the dipole-dipole interactions between various protons as a result of slow molecular motion^{1,2}. Two pulsed nuclear magnetic resonance (NMR) studies, however, have proposed internal magnetic field gradients as the source of spectral broadening. Kaufman *et al.* concluded that these gradients led to inhomogeneity broadening³, whereas Hansen and Lawson proposed molecular diffusion through them⁴. These conclusions were based on the apparent dependence of the spectral width on the applied magnetic field⁵ as well as abrupt changes in transverse relaxation time (T_2) without concomitant changes in the longitudinal relaxation time (T_1) at the bilayer phase boundaries^{4,5}. The dependence of T_2 on pulse spacing in a Carr-Purcell experiment⁶ has also been taken as evidence for molecular diffusion through internal magnetic field gradients⁴.

We wish to report NMR relaxation measurements to support the interpretation that the NMR spectral widths of lipids in the bilayer phase arise from slow molecular motion. We have made continuous wave (CW) as well as pulsed NMR measurements on lecithin bilayers dispersed in D_2O . Lecithin was purified by the method of Singleton *et al.*⁷, and bilayers were prepared by sealing measured amounts with D_2O in a constricted tube in a vacuum and by centrifuging the mixture back and forth through the constriction. T_1 and T_2 were measured on a Bruker B-KR 321 pulsed NMR spectrometer at 30° C. The CW spectrum of lecithin was obtained on a Varian HR-220 spectrometer at 18° C. The Fourier transformed spectrum was recorded at 30° C with a Varian HA-100 spectrometer and Fourier transform accessory interfaced to a Varian 620i computer.

The log of the free induction decay against time in a magnetic field of 14.1 kgauss for the protons of a lecithin bilayer sample is shown in Fig. 1. The free induction decay is clearly non-

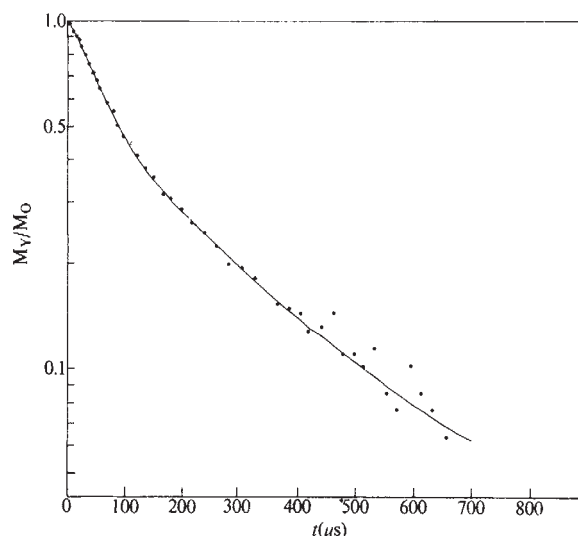


Fig. 1 The proton free induction decay of a lecithin bilayer sample at 30° C in a magnetic field of 14.1 kgauss.

exponential, with the signal decaying to $1/e$ of its original value in 135 μ s, which can be considered the effective transverse relaxation time, T_2^{eff} , for the system. Examination of the signal with a 90° - 90° pulse sequence⁸ showed that the signal contained no component with T_2 much shorter than T_2^{eff} . Because of sensitivity limitations, it was not possible to examine the free induction decay beyond ~ 700 μ s. It was apparent, however, that the signal appearing during the first 500 μ s included the bulk of the magnetization of the sample.

T_1 was measured using the standard 180° - 90° pulse sequence⁶. In Fig. 2 we have plotted the log of the recovery of the magnetization after the 180° pulse against the spacing between the 180° and 90° pulses, which shows that the time dependence of the recovery of the magnetization is exponential over a period of 5 T_1 s, with $T_1 = 220$ ms. The longitudinal relaxation thus seems to be described by one characteristic time only.

The high resolution PMR spectrum of lecithin bilayers at 220 MHz is shown in Fig. 3. For comparison we have also depicted the Fourier transformed spectrum of the same sample recorded at 100 MHz. In the Fourier transform experiments, the spectrometer receiver was switched out for 250 μ s after each pulse in order to avoid receiver saturation. Thus, any broad component, characterized by $T_2 < 250$ μ s or a linewidth $> \sim 1,000$ Hz, should be missed by the receiver and thus not contribute to the Fourier transformed spectrum. The latter is therefore best thought of as a high resolution spectrum, in which components with linewidths in excess of several thousand cycles are filtered out instrumentally. Thus, whereas the pulsed experiments monitor those protons with short T_2 values, the high resolution Fourier transformed spectrum yields information about those protons with long transverse relaxation times only. In spite of the difference in the appearance of the two spectra, the three resonances which do appear in the Fourier transformed spectrum can be correlated with those in the 220 MHz CW spectrum. The resonance which appears at 1.6 p.p.m. upfield from HOD can be assigned to the choline methyls on the ionic head of the lecithin molecule. The highest field resonance (at 3.9 p.p.m. upfield from HOD) results from the terminal methyl groups of the hydrocarbon chains. The shoulder on the low field side of this resonance represents a contribution from the methylene protons.

If the results are to be interpreted in terms of internal magnetic field inhomogeneities or molecular diffusion through internal magnetic field gradients, we would expect the effective transverse relaxation time to depend on the magnetic field. For this reason we have measured T_2^{eff} at 3.76, 8.92 and 14.1 kgauss (Table 1). It is significant that T_2^{eff} is independent of

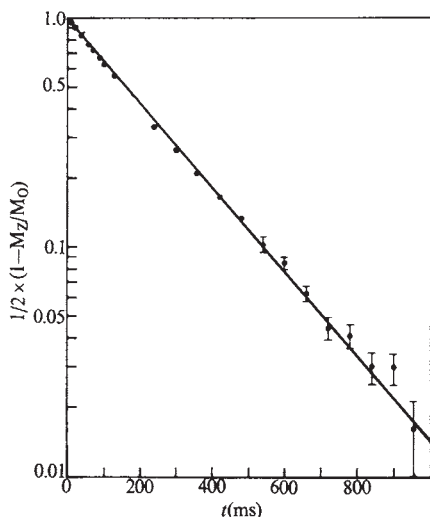


Fig. 2 Recovery of the magnetization of the protons in a lecithin bilayer sample after a 180° pulse at 30° C in a magnetic field of 14.1 kgauss.

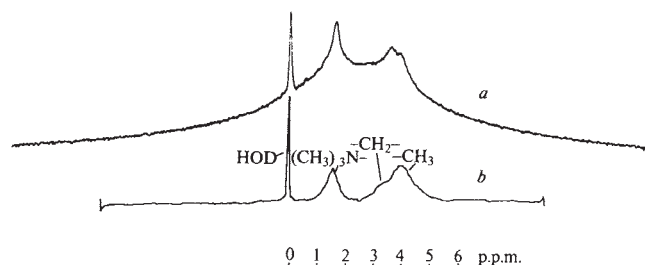


Fig. 3 High resolution proton magnetic resonance spectra of a lecithin bilayer sample: (a) 220 MHz spectrum at 18° C; (b) 100 MHz Fourier transformed spectrum at 30° C.

the applied magnetic field, within experimental error. This result, we believe, rules out internal magnetic field gradients as the primary cause of the breadth of the PMR spectrum.

The non-exponential behaviour of the free induction decay, as well as the line shape of the CW spectrum, suggest strongly that there is a distribution of transverse relaxation times for the various protons in the lecithin bilayers. This interpretation is confirmed by the Fourier transform results. The intensities of the various resonances in the Fourier transformed spectrum indicate that a significant fraction, if not all, of the terminal methyl protons of the hydrocarbon chains have a transverse relaxation time longer than 250 μ s, whereas this applies only to a small percentage of the methylene protons. A comparison of the CW and the Fourier transformed spectra further suggests that there is an abrupt increase in the transverse relaxation time near the end of the hydrocarbon chain. If this were not so, a broad methylene component should be superimposed on the narrower methyl and methylene resonances in the Fourier transformed spectrum.

Table 1 Magnetic Field Dependence of the Transverse Relaxation Rates

Magnetic field (kgauss)	$1/T_2^{\text{eff}}$ (s^{-1})
3.76	$6,900 \pm 500$
8.92	$6,700 \pm 500$
14.1	$7,400 \pm 500$

Because, in the T_1 experiments, the magnitude of the magnetization was measured near the peak of the free induction decay, the T_1 value which we obtain is a single T_1 for all the protons, including the methylene protons. These account for 72% of all the protons in egg lecithin⁷, and have the very short T_2 values observed. We believe that the observation of one longitudinal relaxation time for all the protons of the hydrocarbon chain implies that only a small number of these protons are significantly coupled to the lattice. When the correlation time for molecular motion is long compared with the reciprocal of the Larmor precession frequency, T_2 will be much shorter than T_1 , and spin coupling between protons is more efficient than thermal relaxation⁹. Spin diffusion can then permit the magnetization to travel to the appropriate heat sinks from regions which are weakly coupled to the lattice¹⁰. That is, because of the efficient spin-spin coupling between the protons, thermal relaxation can only occur through those protons most effectively coupled to the lattice. In the bilayer phase, we expect the end groups of the hydrocarbon chains to be the most mobile part of the molecule, and therefore the heat sinks to be located in this region^{11,12}. It is possible that the terminal methyl groups are most efficiently coupled to the lattice. But if these end groups are sufficiently mobile, the methyl protons may not be as efficiently coupled to the lattice as some methylene protons further up the chain. Therefore, the measured value of T_1 only gives information about that part of the hydrocarbon chain which serves as the heat sink.

From these results it seems appropriate to comment on the Carr-Purcell results^{3,4}. Because of the time scale on which

these pulse sequence measurements were made, it is clear that the greater part of the free induction decay in these experiments would be lost by the time of the first Carr–Purcell pulse, and thus only the tail of the decay could contribute to the echo signal. These Carr–Purcell results therefore do not provide insight into the dominant relaxation mechanism.

To sum up, nuclear relaxation measurements can elucidate the state of molecular motion in bilayer phases. In the case of lecithin bilayers, we have shown that the motion of the hydrocarbon chain is sufficiently slow to lead to incomplete averaging of dipole–dipole interactions. But it seems that the end of the hydrocarbon chain is significantly more mobile than the rest of the chain.

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Radiocarbon Dating of Proto-Solutrean in Wales

IN his report on radiocarbon evidence of the age of the Irish Sea Glaciation in the Vale of Clwyd¹, Rowlands expressed surprise that the radiocarbon dating of a mammoth carpal from deposits in Ffynnon Beuno cave containing "Aurignacian and Proto-Solutrean artefacts" was only $18,000 \pm 1,400$ – $1,200$ yr BP (Birm.–146); because, on the basis of comparison with the radiocarbon ages of such industries in France, the dating was expected to be 10,000 yr older. I believe that he will be reassured to hear that a fossil bone found in the same archaeological context at another Welsh site proved to be of almost identical antiquity. In 1968, the radiocarbon age of collagen extracted from the skeleton of Paviland Man, preserved in the University Museum, Oxford, was measured by Richard Burleigh in the British Museum Research Laboratory, and the result he obtained was: $18,460 \pm 340$ yr BP (BM–374). In reporting this date of the "Red Lady of Paviland"² I pointed out that the records of excavation at the site (Goat's Hole) showed that the human skeleton was associated with remains of mammoth and with Upper Palaeolithic artefacts including types identified by D. A. E. Garrod as Final Aurignacian and Proto-

Solutrean. Some archaeologists were surprised that the Paviland dating was not higher, but I was sufficiently sure of its reliability to infer provisionally that in South-West Britain, Proto-Solutrean points and Aurignacoid artefacts were being made long after they had been superseded in France. In the forthcoming *Catalogue of Fossil Hominids*, Part 2 (Europe), I have therefore indicated the cultural horizon of Paviland Man as late Proto-Solutrean.

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BIOLOGICAL SCIENCES

Environmental Fluctuations and Population Size

BOROWSKY has analysed the magnitude and pattern of the temporal fluctuations of experimental populations of *Drosophila*¹, and has shown that the fluctuations are not randomly distributed in their direction; the frequencies of the competing species change in the same direction (upwards or downwards) more frequently than would be expected by chance alone. Moreover, he has demonstrated that the fluctuations are larger in amplitude than is expected in a binomial distribution. He concludes correctly that some external factor or factors are responsible for the magnitude and non-random association of the fluctuations, on which the most important environmental influence is probably temperature. I believe, however, that other uncontrolled variations in the quality and quantity of food, humidity, and so on, may have contributed as much as, or more than, temperature to the pattern and amplitude of the fluctuations of the experimental populations.

At 23.5° C *Drosophila serrata* and *D. pseudoobscura* coexist in competitive conditions in laboratory populations². The average number of flies of both species in the mixed populations is considerably less than their mean number in single species populations. Borowsky contends that this difference can be accounted for if the experimental temperature fluctuates between 23° and 24° C. I have plotted in Fig. 1 the number of flies, in the ordinate, against temperature, in the abscissa. If *a* represents the mean number of flies at 23° C and *b* the mean number of flies at 24° C, and if temperature fluctuates between 23° and 24° C with a mean of 23.5° C, and the function relating number of flies to temperature is linear, then the average number of flies with oscillating temperature is *c*, the same as the number of flies at the mean temperature. If the function is convex the number of flies at the mean temperature (*d*) will be smaller, and if it is concave (*e*) it will be greater, than the average number of flies in a fluctuating environment. These conclusions can be extended to any other fluctuating environmental variables.

Borowsky assumes that the number of flies is a linear function of temperature. In that case, the average number of flies with fluctuating temperature will be the same as the number at the mean temperature. Changes in temperature will result in a change in the frequency of the species. When the frequency of one species decreases, the numbers of that species decrease and those of the other species increase. Borowsky points out only the decrease in the numbers of one of the species. If changes in frequency are accomplished exclusively by a decrease in the numbers of one of the species, then tempera-

ture fluctuations would lead to a rapid decrease in the total number of flies. The number of flies at a given mean temperature, however, remains approximately constant throughout the experiment^{2,3}.

Gause and Witt^{3,4} have studied the conditions which may lead to the stable coexistence of two competing species. The instantaneous rate of growth of two species growing in the same limited environment is given by the equations

$$\begin{aligned}\frac{dN_1}{dt} &= r_1 N_1 \left(1 - \frac{N_1}{K_1} - \frac{\alpha N_2}{K_1} \right) \\ \frac{dN_2}{dt} &= r_2 N_2 \left(1 - \frac{N_2}{K_2} - \frac{\beta N_1}{K_2} \right)\end{aligned}\quad (1)$$

where r is the innate rate of increase of the species, N is the number of individuals at a given time, K is the saturation density of the species when only one species is present, α and β are the coefficients of competition, and the subscripts 1 and 2 refer to the two species. Gause and Witt conclude that a stable coexistence between two competing species obtains only if

$$\alpha < \frac{K_1}{K_2} \quad \text{and} \quad \beta < \frac{K_2}{K_1} \quad (2)$$

This case has been represented in Fig. 2. The necessary conditions for a stable equilibrium (2) imply that the numbers of the species at equilibrium (a) are above the straight line joining K_1 and K_2 .

Table 1 gives the mean number of flies in the competing populations² at 23.5° C (weeks 6–23) and at 23° C (weeks 29–40). I have measured the effect of temperature on population size in single species cultures of *D. serrata*⁵ and *D. pseudoobscura*⁶. Assuming that the temperature has a linear effect on population size, a decrease of 0.5° C will result in an increase of 2.3% in the numbers of *D. serrata* and of 6.3% in the numbers of *D. pseudoobscura*. Lack of linearity in these single species populations is not likely to affect substantially the argument to be presented below. I have used these estimates to calculate the mean number of flies in single species cultures at 23° C from the observed means at 23.5° C.

The observed frequencies of the two species at equilibrium can be used to estimate the lower bound of the number of both species expected from equations (1). If the inequalities (2) obtain, the lower bound of the number of the two species is

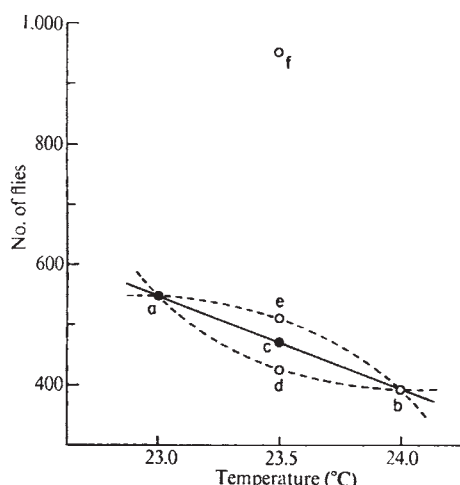


Fig. 1 The relationship between population size, ordinate, and temperature, abscissa. The two solid circles, a and c , represent the observed total number of flies of two species, *Drosophila serrata* and *D. pseudoobscura* AR, at 23° and 23.5° C respectively. b is the estimated number of flies at 24° C; d and e represent the number of flies at 23.5° C when the function relating number of flies to temperature is convex or concave, respectively. To satisfy the Volterra–Gause equations, the total number of flies of both species should be above f .

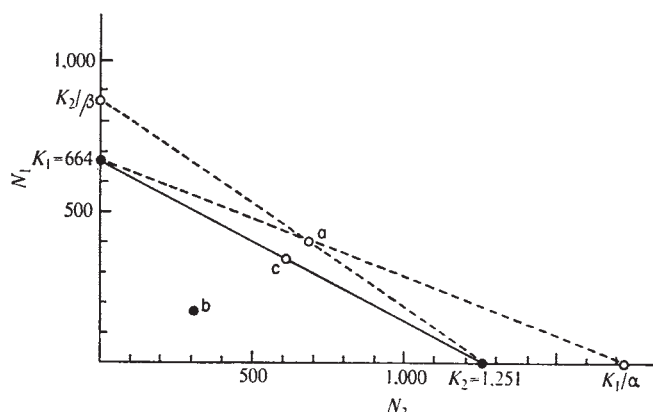


Fig. 2 The Volterra–Gause equations predict that a stable equilibrium will exist only above the straight line joining K_1 and K_2 , that is, when

$$\alpha < \frac{K_1}{K_2} \quad \text{and} \quad \beta < \frac{K_2}{K_1}$$

b specifies the observed numbers of *Drosophila serrata* and *D. pseudoobscura* AR at 23.5° C. For the observed equilibrium frequencies of the two species the Volterra–Gause equations predict that a stable equilibrium can exist only at a point, a , which is above c .

defined by the points lying in the line joining K_1 and K_2 . The conditions (2) require that at equilibrium the numbers of each species be above such line. The mean numbers of flies in the competition populations are in reality substantially lower than the lower limit of equilibrium numbers predicted by the equations (1). In Fig. 2, the solid circle, b , indicates the observed numbers of the AR populations at 23.5° C; c represents the expected numbers as given in Table 1. The numbers specified by c are smaller than the numbers, a , at which a stable equilibrium is possible according to equations (1). In Fig. 1 the two solid circles, a and c , represent the observed number of flies of both species in the AR populations at 23° C and 23.5° C respectively. The number of both species at which a stable equilibrium at 23.5° C is possible should be, according to the conditions (2), above the open circle, f . The differences between the observed and the expected numbers are so large that it seems safe to conclude that small fluctuations in temperature, or in any other environmental factor, cannot account for the decrease in the number of flies observed at equilibrium relative to the numbers which would satisfy conditions (2).

In my earlier communication², I gave the temperature at which the experiments were conducted as 23.5° ± 0.5° C. The manufacturer of the incubator used for the experiment guarantees that temperature fluctuations will not exceed 0.5° C above or below the specified temperature. I have thermo-

Table 1 Mean Number of Flies of *D. serrata* and *D. pseudoobscura* in Pure and Mixed Cultures

Species	23.5° C	23° C
Single species populations:		
<i>D. serrata</i>	1,251	1,280 *
<i>D. pseudoobscura</i> AR	664	706 *
<i>D. pseudoobscura</i> CH	603	641 *
Mixed populations:		
<i>D. serrata</i>	303	201
<i>D. pseudoobscura</i> AR	167	346
Observed total	470	548
Expected total †	> 952	> 845
<i>D. serrata</i>	563	463
<i>D. pseudoobscura</i> CH	78	223
Observed total	641	685
Expected total †	> 1,106	> 966

* Estimated from the means at 23.5° C.

† Expected equilibrium values to satisfy the Volterra–Gause equations.

graphs recording the temperature continuously throughout the experimental period. The temperature has fluctuated between 0.1° and 0.2° C of the specified value, in cycles of 20–30 min.

Recent experiments have confirmed that the competitive fitness of *Drosophila* species is frequency dependent⁷. A stable equilibrium obtains in the experimental populations because above its equilibrium frequency a species has lower fitness, and below its equilibrium frequency higher fitness, than the competing species. The two species compete for the same limited resources and, moreover, there are other mutual negative interactions which depress their numbers in the mixed populations.

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Deleterious Mutations and Neutral Substitutions

CLARKE¹ says that his observations "invalidate" several "less critical" studies, in which category he places one by us². Clarke's calculations are based essentially on two contributions by others; these are the "objective classification of the chemical properties of the amino-acids" by Sneath³ and two tables in Dayhoff's *Atlas*⁴. We believe that classification procedures for amino-acids should be used with caution. Indeed, Clarke quotes a remark by Zuckerkandl and Pauling⁵ that "apparently chemists and protein molecules do not share the same opinions regarding the definition of the most prominent properties of a residue". There is much point to this remark. In the few large families of homologous proteins that have been studied (haemoglobins, cytochromes and immunoglobulins), many sites are occupied by six or more different amino-acids². This implies, as is well known, that a great deal of interchangeability is possible at such sites. The flexibility of interchange may reflect a lack of close proximity to other residues in the tertiary structure of the protein.

The table by Dayhoff (ref. 4, page 76, Fig. 9–3) was constructed on the basis of "simplified phylogenetic trees", made by comparing the sequences of proteins. Dayhoff states, "In practice some of the positions in the nodal sequences are blank (ambiguous). For these, we have treated the changes statistically, distributing them among all observed alternatives"⁴. Haemoglobin chains were a part of the information used by Dayhoff in constructing her Fig. 9–3. The sequences of some regions of these were inferred by homology and this procedure tends to hide the very information being sought. Dayhoff's table therefore presents certain problems of interpretation.

It was clearly Dayhoff's aim to tabulate only individual evolutionary events, not all the possible amino-acid residues that could occur at a specific locus. Clarke has utilized only substitutions referable to single base changes. But many of the tabulated amino-acid differences in extant sequences may be due to an accumulation of repeated changes at a given site which are only coincidentally referable to single base changes. Thus Clarke's data consist of an unknown mixture of single evolutionary steps and of the end results of multiple evolutionary steps. This is not too serious an objection, because what we are really interested in is the number of different amino-acids that can occupy a given site in a functional

protein. But if this is the case, it seems more profitable to consider all of the amino-acid substitutions, referable to single base interchanges, that can occur at a given site.

The haemoglobin and myoglobin chains are all homologous; indeed, this family of proteins is one of the classical examples of homology⁶; because their tertiary structures are well established, it is possible to align the sequences of the globins so that corresponding sites are matched against each other. A comparison of the amino-acid residues with each other at each homologous site indicates that sixty-five of the seventy-five possible interchanges corresponding to single base codon changes may have occurred in the globin series.

Clarke states that he could, "without previous knowledge of protein chemistry, have predicted the importance of the disulphide bond". He reaches this conclusion on the basis of the rarity of single-base interchanges involving cysteine. Many of the cysteine residues in the proteins forming the basis of Dayhoff's table are in cytochrome *c* (twenty-five examples in the *Atlas*), in ferredoxin (seven examples), and in α and β haemoglobins and myoglobin (thirty examples). These proteins do not contain disulphide bonds. Two cysteines in each cytochrome *c* are in covalent linkages with haem; they are invariant and hence do not participate in evolutionary interchanges. There are serious statistical biases in the data in Dayhoff's Fig. 9–3. Some families of proteins are represented by many examples: cytochrome *c*, the globins and insulin. Others have only minimal representation. The relatively small (about fifty residues) insulin molecule contains six cysteines, and is represented in the *Atlas* by fourteen examples. The cysteines are invariant, but most of the entire molecule is invariant in most of the species studied. Thus this one family heavily weights the data on cysteine.

The importance of disulphide bonds in proteins such as insulin was discovered by protein chemists in the course of obtaining the information used by Clarke. His paean, that "it is satisfying that the student of evolution can perhaps contribute to solving the problems of the chemist"¹, is therefore not borne out by his article.

Clarke concludes that the "apparent correspondence" (to the Poisson distribution of substitutions for several proteins, excluding invariant sites) "reported by King and Jukes is an accident"¹. Actually, we used the Poisson distribution primarily to draw attention to the fact that many so-called hyper-variable sites "are predictable in terms of the Poisson distribution"². We prefer a model stating that point mutations occur randomly in DNA, and therefore in proteins, but the adaptive process screens out deleterious mutations. Evolutionary replacements will therefore deviate from the Poisson distribution, but a certain partial or residual randomness is perceptible in such replacements when proteins which have considerable latitude for variation are studied. We have not claimed either that all variable sites are equally variable or that such a condition would be a prerequisite for non-Darwinian evolution.

In spite of these reservations, we find Clarke's analysis to be an interesting contribution to molecular evolution. If his Fig. 1 is taken at face value, it is good substantiation for the concept of a predominance of neutral evolutionary changes, for it provides further evidence that evolutionary substitutions are correlated with the chemical similarity of amino-acids.

To be operationally neutral, a mutated allele need not be functionally identical to the original form. Drift effects will predominate over selection effects if the selection coefficient is within $\pm 1/2N_e$ of absolute neutrality⁷. The more similar two amino-acids are, the greater will be the probability that substitution of one for the other will not result in a significant change in the function of the protein. If selectively neutral and nearly neutral changes are common in evolution, one should expect to find exchanges between chemically similar amino-acids to be favoured over those between dissimilar amino-acids.

In contrast, positive Darwinian evolution requires that there

must be a difference in the function of the changed protein (or gene) that can be detected by the organism. Only a small percentage even of favourable mutations will be successfully incorporated as evolutionary changes; and the probability of success is directly proportional to the magnitude of the selective advantage. Thus a selectionist hypothesis might predict radical changes to be favoured over conservative changes.

Clarke extended the slope of the regression in his Table 1 to intercept the point of zero dissimilarity, thus giving an expectation of the relative probability of evolutionary substitution for absolute selective neutrality. This computed expectation is some eleven times the average observed rate of substitutions between different real amino-acids, and Clarke concluded that "at least 85–90% of the mutations causing a change in amino-acid are, on the average, selectively disadvantageous . . . King and Jukes have estimated that 5–10% of mutations are neutral. At first sight my own calculations seem to support them. . . ." Here we are quite in agreement and Clarke's calculations do corroborate ours. Unfortunately, he then proceeds to ignore his own calculations, for he continues ". . . but they do not. [The calculations] take into account only one kind of selective constraint, and there are many others . . . the kind of protein . . . the position in the protein . . . the type of organism. . . . When all such factors are taken into account it seems likely that our estimate of the average proportion of 'neutral' mutations will be nearer 0.1%." No basis is given for this guess, and the statement makes no sense at all to us. All these "other" selective constraints were certainly operating in the evolutionary events which gave rise to the data on which his calculations were based. Ultimately all constraints on interchanges between two amino-acids depend on the chemical differences between them.

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Antitrachoma Activity of Rifamycin B and 8-O-Acetylrimfamycin S

It has been found that rifampicin¹ inhibits^{2–4} irreversibly^{5,6} the development of trachoma agent, and that the effect of rifampicin on trachoma is caused by the inhibition of the agent's DNA dependent RNA polymerase present in the trachoma elementary bodies^{7,8} and in the developing inclusion bodies. Thus, rifampicin inhibits trachoma agent and prokaryotic cells by the same mechanism⁹. Studies have shown⁴ that the presence of the hydrazone side chain in the rifamycin SV molecule enhances the antitrachoma activity of the antibiotic. We investigated the antitrachoma activity of rifamycin B, a natural fermentation product of *Streptomyces mediterranei*, and two other rifamycin derivatives which lack anti-RNA polymerase activity¹⁰: compound VIII (23-dehydroxy-27-

demethoxy-23,27-epoxyrifamycin SV) and 8-O-acetylrimfamycin S. We found that only 8-O-acetylrimfamycin S effectively inhibited the development of trachoma agent in FL cell cultures.

The T'ang strain of trachoma agent (TRIC/RC-PK/PK-2/OT) was propagated in FL cells. Compound VIII, 8-O-acetylrimfamycin S and rifampicin B¹⁰ was obtained from Ciba Ltd, Basle, and rifamycin SV and rifampicin¹ from Gruppo Lepetit, Milano. The antibiotics were dissolved and, at concentrations ranging from 0.001 to 100 $\mu\text{g ml}^{-1}$, added to the trachoma-infected FL cultures, which were then incubated at 37° C for 48 h. We determined the smallest concentration of the antibiotic that completely inhibited the development of trachoma inclusion bodies.

Table 1 Effect of Rifamycins on the Development of Trachoma Agent in FL Cell Cultures

Compound	Inhibition of RNA polymerase	Minimal anti-trachoma inhibitory dose ($\mu\text{g ml}^{-1}$)*
Rifamycin SV	+	0.10
Rifampicin	+	0.01
Rifamycin B	+ †	5.0
23-Dehydroxy-27-demethoxy-23,27-epoxyrifamycin SV (VIII)	– †	No effect at 10.0‡
8-O-acetylrimfamycin S	– †	0.10

* FL cells were infected with the T'ang strain of trachoma agent. To the infected cultures various concentrations of the rifamycins were added and the cultures were incubated at 37° C. After 48 h the cultures were scored for the appearance of specific trachoma inclusion bodies. The minimal dose which completely inhibited the development of trachoma inclusion bodies is given.

† Based on the findings reported by Wehrli and Staehelin¹⁰ on the interaction between rifamycins and bacterial RNA polymerase.

‡ At a concentration of 100 $\mu\text{g ml}^{-1}$ the antibiotic exerted a toxic effect on the cells. At 10 $\mu\text{g ml}^{-1}$ trachoma inclusion bodies were observed.

The results (Table 1) demonstrated that rifamycin B had a low antitrachoma activity (5 $\mu\text{g ml}^{-1}$) as compared with rifamycin SV (0.1 $\mu\text{g ml}^{-1}$) and rifampicin (0.01 $\mu\text{g ml}^{-1}$). Of the two rifamycins which lack anti-RNA polymerase activity¹⁰ compound VIII had no effect on the development of the trachoma agent. At 10 $\mu\text{g ml}^{-1}$ there was no observable effect and higher doses were toxic to the cells. In contrast, 8-O-acetylrimfamycin S effectively inhibited the development of trachoma agent in FL cells at a concentration of 0.1 $\mu\text{g ml}^{-1}$. Injection of 8-O-acetylrimfamycin S into trachoma infected embryonated eggs³ had no effect on the course of infection, even at a concentration of 100 μg per egg. Rifampicin was inhibitory in infected embryonated eggs at a concentration of 10 μg per egg.

Therefore 8-O-acetylrimfamycin S can inhibit the development of trachoma agent in FL cells. Because this compound cannot inhibit bacterial RNA polymerase, we assume that trachoma agent is inhibited by a mechanism so far unknown. As 8-O-acetylrimfamycin is not very stable in alkaline or neutral conditions the activity may be caused by the gradual release of rifampicin S (Bickel and Staehelin, personal communication). But we found that inhibition of trachoma development starts with the addition of the antibiotic.

The possible use of rifampicin in the treatment of human trachoma patients could be complicated by the appearance of trachoma mutants resistant to rifampicin (E. Jawetz, personal communication). It is therefore essential to search for rifamycin derivatives that will inhibit the trachoma agent by different mechanisms, which do not involve the inhibition of the agent's RNA polymerase. We have demonstrated here that 8-O-acetylrimfamycin S might have a new mode of action, and a study of more rifamycin derivatives that lack anti-RNA poly-

merase activity could lead to the isolation of effective rifamycins, which might have two different modes of antibacterial activities. New rifamycin derivatives have been reported¹¹ that inhibit the growth of the bacterial mutants resistant to rifampicin.

I thank Dr Zichriya Zakay-Rones, Mrs Yael Asher, Karla Press and Nehama Himmel; also Professor M. Staehelin and Dr H. Bickel of Ciba Ltd for rifamycin B, 8-O-acetyl-rifamycin S and compound VIII, and Professors P. Sensi and L. Silvestri of Gruppo Lepetit for the rifampicin and rifamycin SV. This work was supported by grants from the World Health Organization and the National Institutes of Health, Bethesda, Maryland.

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Immunological Implications of Ultrastructural Studies of Goat × Sheep Hybrid Placentae

THERE is ample evidence that conception takes place when female goats are inseminated with sheep semen, and that the resulting hybrid embryo fails to survive beyond the second month of gestation^{1,2}. The cause of embryonic death has not

been determined but previous studies have shown that all cases are associated with certain gross and histological abnormalities of placental structure, reflected in failure either to establish or to maintain a normal placental relationship^{3,4}.

We present the results of an investigation designed to obtain more details of the nature of the pathological changes in the hybrid placenta and of the order in which they occur. Hybrid pregnancies were established by insemination of goats with sheep semen and normal pregnancies were obtained by natural mating. Tissues of animals under pentobarbitone sodium ('Nembutal'; Abbott Laboratories Ltd) anaesthesia were fixed *in situ*. The uteri were exposed at laparotomy and the allantoic fluids were replaced with Karnovsky's formal-glutaraldehyde fixative⁵. After approximately 5 min a lethal dose of 'Nembutal' was administered and the uterus was removed. The uterus was incised and fixation was continued for a further 2 to 4 h at 4° C. Blocks of tissue were prepared and, after overnight fixation, 250 µm slices were cut on a Sorval tissue chopper and embedded in 'Araldite'.

After 34 days the ultrastructure of the hybrid placenta corresponded broadly with the macroscopic features and with the features seen by light microscopy. The maternal crypts and the foetal villi were shallower than normal but the trophoblast was firmly attached to the uterine epithelium, and there was evidence of active cell division in the trophoblast. Immediately beneath the uterine epithelium of the placentomes, however, the maternal blood vessels showed the presence of large numbers of platelets, either alone or mixed with isolated red blood cells. In most cases the platelets were loosely aggregated and were of normal shape; few were degranulated, and no fibrin strands were present (Fig. 1a). Platelets were found neither in the maternal blood vessels of the deeper part of the placentome nor, at this stage, in the sub-epithelial vessels in the inter-placentome regions. No platelets were observed in the foetal blood vessels. Counts were made of the numbers of vessels with and without platelets in thick 'Araldite' sections examined by light microscopy. At this stage platelets were seen in only 2 of 47 maternal vessels examined in normal placental tissues; by contrast, fifty-seven out of seventy-five vessels in the hybrid placenta contained platelets, and twenty-four of these contained platelets exclusively.

In the 38 day hybrid placenta, platelets were prominent in the sub-epithelial vessels of the inter-placentome tissue. Thirty-seven out of fifty-five vessels contained platelets, and fourteen contained platelets exclusively. In normal goat placenta 148 vessels were examined from inter-placentome areas and none contained platelets. The sub-epithelial vessels in the placentomes of the 38 day hybrid placenta did not contain

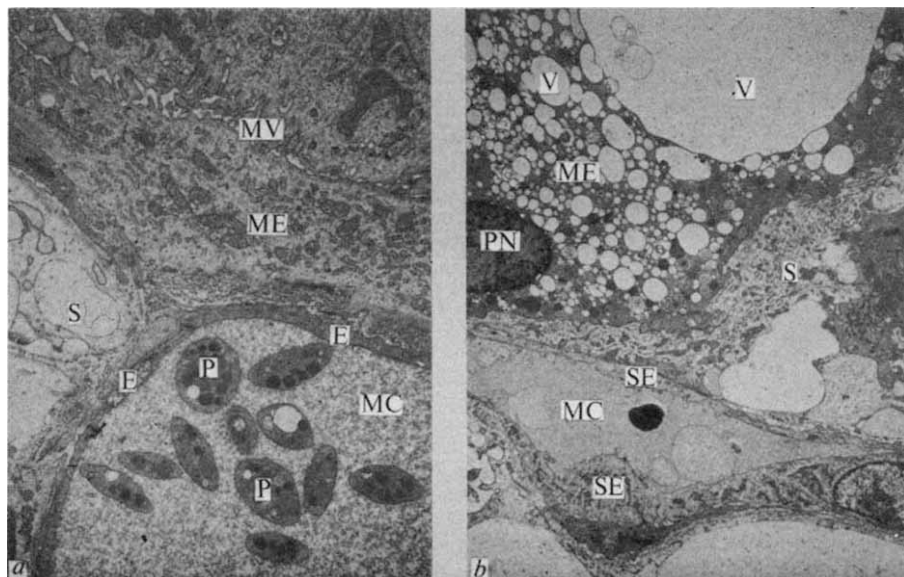


Fig. 1 Ultrastructural features of the goat × sheep hybrid placenta. a, Placentome, 34 days gestation, showing platelet aggregation in maternal blood vessel (×5,000). b, Placentome, 38 days gestation, showing degenerative changes in the vascular endothelium and uterine epithelium (×3,000). E, Endothelium; MC, maternal sub-epithelial capillary; ME, uterine epithelium (endometrium); MV, microvilli; P, platelet; PN, pyknotic nucleus; S, stroma; SE, swollen endothelium; V, vacuole.

platelets but had lobular swellings of the endothelium (Fig. 1b) which in some cases obliterated the lumen. In some areas the endothelium had become discontinuous and haemorrhage had occurred. Degenerative changes were also evident in the uterine epithelium which had a lace-like appearance caused by extensive vacuolation; in some areas it was completely necrotic. Accumulation of blood between the maternal tissues and the trophoblast occurred in areas where both endothelium and uterine epithelium had broken down. At this stage there was no necrosis of inter-placental tissue.

No evidence of platelet aggregation, of swelling of the endothelium or of degenerative changes in the uterine epithelium was found either in the placentomes or in the inter-placental regions of normal goat placentae examined after 27, 34 or 39 days of gestation.

At the stages examined hybrid trophoblast was structurally normal; the evidence points to the involvement of maternal tissues in the initial damage to the placenta. The nature of the lesions supports the suggestion that death of the hybrid can be explained in immunological terms³. Platelet aggregation and lobular swelling of the endothelium of blood vessels are associated with injury caused by antigen-antibody complexes⁶. Changes in the endothelium resembling those described here have been found in transplanted hearts and are said to be characteristic of the changes which occur in visceral homographs⁷. Platelet aggregation also occurs in the blood vessels of kidneys transplanted to dogs previously sensitized by skin grafts⁸.

Antigen-antibody complexes on the maternal side of the placenta could derive either from a reaction by the hybrid foetus against maternal antigens or from a reaction by the mother to foetal antigens. The second possibility is made the more likely by observations that the life of the hybrid embryo is shortened in goats which have had a previous hybrid pregnancy³ and in goats which had previously received skin grafts and injections of leucocytes from the sheep sire (unpublished results of P. T. M.).

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Lack of Correlation between Conversion by RNA Tumour Viruses and Increased Agglutinability of Cells by Concanavalin A and Wheat Germ Agglutinin

ALTERATIONS in the cell surface seem to accompany neoplastic transformation of cells in culture. Such alterations may affect cell orientation and control of cell multiplication. Recently,

interest has focused on the accessibility to various plant glycoproteins of specific receptor sites on the cell surface as a measure of specific changes in the cell surface accompanying transformation induced by various carcinogenic agents, including the DNA tumour viruses¹⁻⁶. We are not, however, aware of any published work dealing with the effects of infection with RNA tumour viruses on the response of cells to such glycoproteins.

The RNA tumour viruses provide a good system for the study of these relationships because the specificity of changes associated with transformation of the infected cells can be studied within a few days of infection, and can be compared with untransformed, non-infected cells and untransformed virus-producing cells. We have looked at agglutination with wheat germ agglutinin and concanavalin A of parallel uninfected, and Rous sarcoma virus (RSV) and avian leucosis virus (RAV)-infected chicken cells; and parallel uninfected, and murine sarcoma virus (MSV) and murine leukaemia virus (MLV)-infected rat and mouse cells, all in secondary or tertiary cultures. Two lines of rat cells converted by RSV were also studied.

Sources of viruses, techniques for cell culture and virus infection were as described before⁷⁻⁹. Prague virus (P-RSV) converted rat cells (XC tumour cells), propagated in tissue culture, were obtained from Dr V. Klement. Cells used in the agglutination test were released from the culture dishes with a 0.02% disodium versenate solution⁶. Some uninfected cells were treated before use in the agglutination test by incubating with 0.01% crystallized, lyophilized trypsin (Worthington Biochemical Corp.) solution in phosphate buffered saline for 2-5 min at 37° C. The trypsin activity was inhibited with soybean trypsin inhibitor, and the cells were then washed three times with phosphate buffered saline. A crude preparation of wheat germ agglutinin was prepared by heating a 10 mg/ml. solution of wheat germ lipase type I (Sigma Chemical Co.) in phosphate buffered saline (Ca²⁺ and Mg²⁺ free) for 15 min at 65° C to inactivate lipolytic activity. The denatured protein was removed by centrifugation at 3,000 r.p.m. for 10 min, and the supernatant, which constituted the crude wheat germ agglutinin, was decanted and stored at -20° C until time of use. Concanavalin A (A grade), lyophilized in NaCl, was obtained from Calbiochem. With the exception of the B77 virus and P-RSV converted rat cells, the agglutination test was performed on all of the cell types about 5 days after infection when morphological conversion became essentially complete in the RSV and MSV-infected cultures. The B77 virus and P-RSV converted rat cells had been propagated in culture for five and more than one hundred passages, respectively. In the agglutination assay serial two-fold dilutions of concanavalin A, with a 1:1 dilution equalling 600 µg/ml. (final concentration equal to 200 µg/ml.), and wheat germ agglutinin, with a 1:1 dilution being the undiluted crude preparation described, were prepared in phosphate buffered saline. Each agglutinin dilution (0.1 ml.) was applied to 0.2 ml. of cell suspension (2-3.0 × 10⁶ cells/ml.) in 10 mm test tubes. After 20 min of incubation at room temperature, 2 ml. of phosphate buffered saline was added to each tube, and the contents were applied to a Petri dish for microscopic observation of the degree of agglutination. A serological scale of 0 to + + + +, based primarily on the cell aggregate size, was used to estimate the degree of agglutination. The approximate sizes of the cell aggregates for the designated degrees of agglutination were as follows: 0, no aggregates; +, two or three cells; + +, three to ten cells; + + +, ten to thirty cells; + + + +, thirty to a hundred cells. The specificity of the agglutinin-cell interaction was demonstrated by agglutination inhibition tests using specific sugar haptenic inhibitors: α-methyl-D-glucopyranoside and α-methyl-D-mannopyranoside for concanavalin A⁶, and N-acetylglucosamine for the wheat germ agglutinin¹⁰.

Results from a typical experiment are plotted in Fig. 1. A summary of all data is given in Table 1. In agreement with

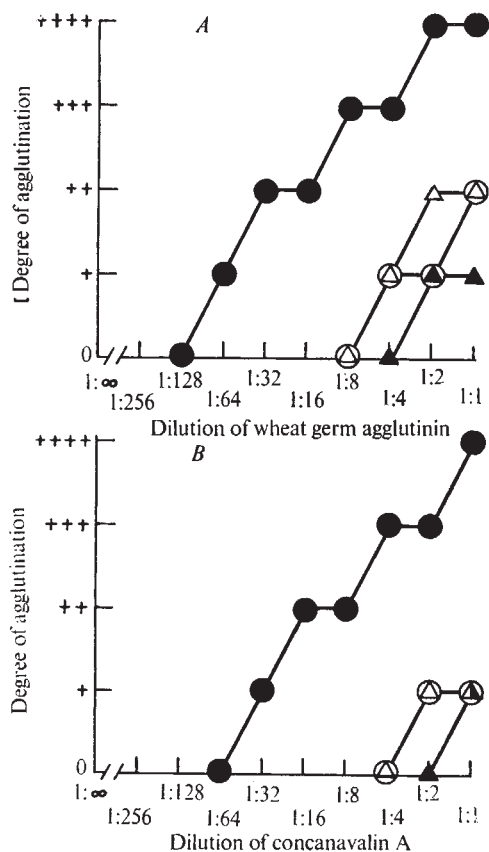


Fig. 1 Agglutination of uninfected, B77 virus and RAV-49 infected chicken cells by wheat germ agglutinin (A), and concanavalin A (B). The agglutination was carried out as described in the text. Uninfected (▲) and trypsinized, uninfected chicken cells (●), B77 infected chicken cells (△), and RAV-49 infected chicken cells (○).

earlier findings^{5,6}, normal cells showed negligible or very low agglutinability. An increase of greater than eight to thirty-two-fold was found after trypsin treatment. With the exception of B77 virus converted rat cells, cells converted by these RNA tumour viruses (Schmidt-Ruppin virus, B77 virus, P-RSV, MSV) did not show marked increases in agglutinability. That

increase which did occur was not specific for conversion, for cells infected by non-converting RNA tumour viruses (RAV, MLV) had essentially the same agglutinability. Rat cells converted by B77 virus, however, displayed markedly increased agglutinability similar to that found in other systems of transformed cells. In control experiments using the conditions described here, we were able to confirm the increased agglutinability of mouse cells transformed by DNA tumour viruses. We used SV3T3 cells supplied by Dr H. Green, and mouse polyoma tumour cells supplied by Dr R. Click.

Our results demonstrate that changes in response to wheat germ agglutinin and concanavalin A are not necessary concomitants of neoplastic transformation in cell culture.

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Table 1 Agglutination of Uninfected, RNA Sarcoma and Leukaemia Virus-Infected Cells by Wheat Germ Agglutinin and Concanavalin A

Cell type	Virus infection	Trypsin treatment	Reciprocal dilution of agglutinin giving ++ degree of agglutination	
			Wheat germ agglutinin	Concanavalin A
Chicken	0	0	<1	<1
Chicken	0	+	32	16
Chicken	B77	0	2	<1
Chicken	RAV-49	0	1	<1
Chicken	SRV*	0	1	ND†
Chicken	RAV-50	0	1	ND
Mouse	0	0	<1	<1
Mouse	0	+	8	8
Mouse	MSV	0	1	2
Mouse	MLV	0	1	4
Rat	0	0	<1	<1
Rat	0	+	16	16
Rat	MSV	0	1	4
Rat	MLV	0	2	2
Rat	B77	0	16	16
Rat	P-RSV	0	2	2

* SRV, Schmidt-Ruppin virus.

† ND, not done.

Survival of Tumours after Immunization against Oestrogens

SOME tumours respond to a circulating hormone by growing at an optimum rate in an abundance of the hormone, or by regressing in its absence¹. Surgical procedures such as ovariectomy, adrenalectomy and hypophysectomy can curtail the growth of these tumours, including those which are susceptible to oestrogens². One such tumour, designated MT/W9A (ref. 3), grows readily in the intact female, regresses after ovariectomy, and grows at optimum rates after the administration of the synthetic oestrogen diethylstilboestrol. These observations, and the finding that antibodies to oestrogens can control the normal physiological responses to oestrogens *in vivo*, formed the basis for our study⁴. We have reported before that actively immunizing sheep against oestrogens by the administration of a conjugate of bovine serum albumin covalently linked to an oestradiol 17 β -hemisuccinate derivative (BSA-E²) resulted in the production of antisera which effectively neutralized the circulating levels of oestrogens; other workers have used a similar approach⁵. We have investigated whether antibodies to oestrogens might induce regression of an oestrogen sensitive tumour.

Female Wistar-Furth rats (about 60 days old) were used because they had been described as suitable hosts for the mammary adenocarcinoma (MT/W9A) which was implanted to the back of each (trochar injection of a 1 mm³ piece). No hormone was administered to any animals. Beginning 28 days after implantation each animal was examined every 3-4 days

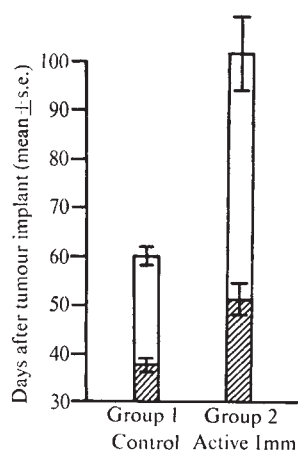


Fig. 1 The effects of active immunization against oestrogens on the onset of tumour growth (hatched area) and survival time (white area) in rats implanted with oestrogen-sensitive tumours.

and the length and width of the tumour were recorded. Selected animals were examined at autopsy for the possible involvement of other organs, although metastasis is seldom noted with this tumour. Both control and immunized animals were tested periodically for any evidence of antibody titre in the peripheral plasma using a technique which has been described as a sensitive and reliable means for detecting antibodies in very low concentration⁵. This is an indirect way of determining the relative titre of antibodies to oestradiol and makes use of the principle with which radioimmunoassays have been developed for the oestrogens. Increasing dilutions of antisera are allowed to equilibrate with 50 pg of tritiated oestradiol for 2 h and the free oestradiol is then removed with dextran-coated charcoal. The dilution of antisera that gives 50% binding of the ³H-oestradiol then gives a relative index of the titre of that antiserum. For a high titred antiserum a dilution of 1/50,000 can often be achieved and still result in the binding of 50% of the ³H-oestradiol.

Twenty rats which served as controls received no treatment, and another twenty were actively immunized against the BSA-E² conjugate once a week for 4 weeks with one booster injection 4 weeks later. Each animal received 1 mg of the conjugate in four divided doses administered subcutaneously in Freund's complete adjuvant and saline. Because the oestradiol was linked to the BSA at the 17 position on the steroid nucleus, the phenolic A ring was available to impart specificity to the antibodies. The antisera produced in several animals had a high degree of specificity to the oestrogens. No other steroid was bound by the antibody to any significant degree. Thus the effects of the active immunization procedures must have been due to the removal of the oestrogens and not to the binding of any other steroid to the antibody.

The two significant findings of this study are shown in Fig. 1. First, the time between implantation and the first evidence of tumour growth was significantly prolonged ($P < 0.001$) from a mean of 36.1 days in the control group to 50.7 days in the actively immunized group. The same relationship between the two groups was found when survival time was determined. The actively immunized animals (group 2) had a highly significant extended survival time ($P < 0.001$) of 100.7 days compared with 60.2 days for the control group. The values for group 2 are reported for the time of writing this communication; several of these animals are still living. In general, the animals which have shown the highest titre of antibodies have survived for the longest period. This is not unexpected and may explain why a few animals in which no antibodies were detected had survival times more like those in the control group.

Because of the relationship between the two significant findings and the level of antibodies detected in the plasma, we are confident that the neutralization of the circulating oestrogens by the antibodies was the primary cause of the extended

survival time and the longer interval between implantation and tumour growth.

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Triple Chromosome Pairing in Triploid Chickens

THE statement that "only two homologous chromosomes can pair at one time" is one of the most basic of cytogenetic principles. It is founded on observations of meiosis in triploid and tetraploid organisms¹⁻⁷ which have suggested that if three homologues are present, they will occur as a pair and a single unpaired chromosome. If four homologues are present, the chromosomes may exchange pairing partners, but they occur as two separate pairs. We now present evidence that the chromosomes of the triploid chicken are not bound by these rules and are capable of "triple pairing".

Triploid chickens occur sporadically. They have the general appearance of hens but have the comb and hackles of a cock and possess a distinct spur on each shank. This tendency to intersexuality is confirmed by the presence of ovotestes. Chromosomally⁸ they are 3A-ZZ. The ovotestes from two such chickens were cut into small pieces in ice-cold Gey's solution and a portion was fixed for thin section electron microscopy. The remainder was pipetted and the resulting single cell suspension was concentrated by centrifugation and allowed to spread on a trough of distilled water. The dispersed cells were picked up on grids, treated with deoxyribonuclease 25 µg/ml. for 5-20 min, and processed as described previously⁹⁻¹¹. This procedure allows the examination of the synaptonemal complexes by electron microscopy.

In the meiosis of diploid organisms, proteinaceous axial cores become attached to the chromosomes during leptotema. During zygonema, homologous axial cores are pulled into a pairing relationship with each other by L-C fibre loops which fuse to form the central element of the synaptonemal complex⁹⁻¹¹. The completed synaptonemal complex, consisting of

two lateral elements (axial cores) and a central element, represents the pairing configuration of homologous chromosomes¹²

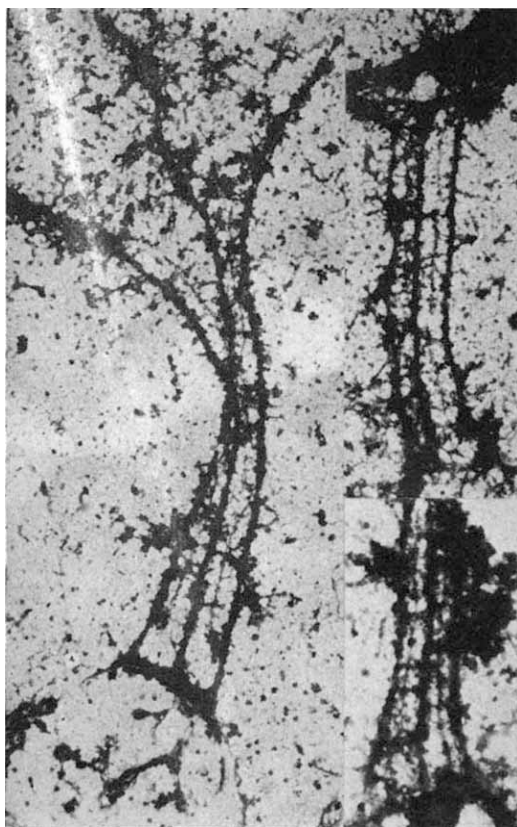


Fig. 1 Pachytene cells of the triploid chicken spread on a water surface, picked up on grids, and treated with DNAase 25 $\mu\text{g}/\text{ml}$. at 20° C for 5–20 min. Three separate triple pairing configurations, consisting of three lateral elements and two central elements, are illustrated. In the left picture the synaptonemal complexes have been pulled apart at the top of the picture. The bottom shows the site on the nuclear membrane to which the lateral elements attach. Similar configurations were not seen in control diploid animals. (Left, $\times 26,160$; upper right, $\times 19,800$; lower right, $\times 26,760$.)

Fig. 1 shows three whole mount preparations of synaptonemal complexes from the triploid chickens treated with deoxyribonuclease. The two synaptonemal complexes complete with central element, side by side, indicate that the three homologous chromosomes have undergone synapsis and completed chromosomal pairing as verified (Fig. 2) by thin section electron microscopy. It was much easier to detect these double synaptonemal complexes in whole mounts than in thin sections. Even in whole mount material not all of the chromosomes showed the double complexes. It was not possible to tell whether the double complexes were present in all chromosomes but became disrupted by the destruction of the delicate L–C fibres and central element during water spreading (as in Fig. 1) or if only a portion of the chromosomes had undergone triple pairing. But as many as five triple paired chromosomes could be observed in one nucleus.

The observation that homologous triple pairing can take place in the triploid chicken has several implications. First, it indicates that there is no pairing force that is suddenly neutralized when two chromosomes pair, thus preventing a third homologue from joining in the union. The question of how the homologous telomeres come to be lined up adjacent to each other at the nuclear membrane^{10,13} during or before the onset of meiosis remains unanswered.

Second, DNA base pairing seems not to be involved in chromosome synapsis. Many lines of evidence, including enzyme digestion studies, suggest that there is no DNA in the

central element, which therefore does not represent the site of molecular pairing of homologous chromosomes^{9–11,14–16}. It is still possible to argue that there may be DNA in the central element that is protected from the enzymes by some unique association with protein, or that DNA is present in such small amounts it cannot be detected, and the central element is the site of molecular pairing; but the observation that three homologous chromosomes may simultaneously be involved in homologous synapsis makes such proposals extremely unlikely. There is no known way for three DNA molecules simultaneously to undergo DNA base pairing, especially when two must be separated from each other by the distance between two central elements. It is, however, conceptually easy to imagine that proteinaceous L–C fibre loops may emanate from the lateral element in two different directions and thus be simultaneously involved in two different central elements.

We have already proposed a relatively simple mechanism of chromosome pairing based on the sequential synthesis, starting at the nuclear membrane, of L–C fibre loops which serially pull together the axial cores into a pairing relationship eventually resulting in the completed synaptonemal complex. The most difficult feature to explain was the multiple switches in pairing partners seen in triploid and tetraploid organisms, because this seemed to require multiple sites (other than the nuclear membrane) of initiation of chromosome pairing. An alternative suggestion is that triple or quadruple pairing can start only from the nuclear membrane. This may then (a) remain as it is, with two or three parallel synaptonemal complexes, (b) spontaneously break down to result in multiple switches in pairing partners, or (c) be broken down by the forces applied in making squash or air dry spreads, and thus only seem to have switches in pairing partners. The latter could not explain all observations for the studies of Moens^{5–7} suggest a switch in pairing partners in the triploid and tetraploid lily in fixed, sectioned material.

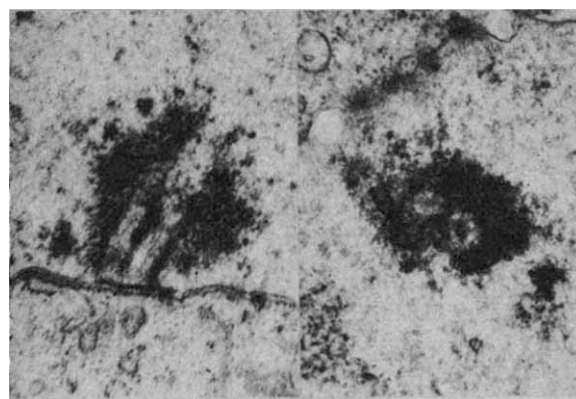


Fig. 2 Thin section electron microscopy of pachytene cells of the triploid chicken. Left, longitudinal section through two adjacent synaptonemal complexes, showing three lateral elements and two central elements ($\times 21,750$). Right, a transverse section of two adjacent synaptonemal complexes ($\times 28,000$). Similar paired synaptonemal complexes were not seen in control diploid animals.

Polycomplexes, which look like a series of stacked synaptonemal complexes, have been described in several organisms¹². The polycomplexes may be arranged side by side or even in a hexagonal array of lateral and central elements. These results are rather embarrassing if one wishes to believe that the central element represents the pairing surface of homologous chromosomes, for these polycomplexes are not associated with the chromatin fibres. It has been suggested that they are aberrant configurations and are not directly related to the synaptonemal complex. Our observation that a lateral element can be involved in pairing in two different directions, however, indicates that it could easily become involved in a serial array of complexes,

and suggests that in a virtual pairing frenzy it could just as easily send out L-C fibre loops in six different directions to form the hexagonal polycomplexes. The polycomplexes may not be so aberrant after all.

It has been suggested that the synaptonemal complex holds certain genes (master genes) in register so that crossing over may occur, and excludes other genes (slave genes) from such recombination^{17-19,7}. If the genes potentially able to participate in recombination are all fixed to a specific portion of the synaptonemal complex, such as the lateral element⁷, the type of triple pairing shown here should place an absolute restriction on recombination between chromosomes 1 and 3 in the absence of intermediary recombination between 1 and 2 and 2 and 3. On the other hand, if recombination takes place in the region around the synaptonemal complex⁹ there should be no such absolute restriction. Investigation of this is somewhat complicated by the fact that homologous chromosomes 1, 2 and 3 probably do not always pair in the same order. Such a problem would be bypassed if a triple crossover event going from 1 to 2, then 2 to 3, and then back to 1 were found. Such crossovers have been described in triploid *Drosophila*²⁰. To vindicate this line of reasoning it will be necessary to demonstrate triple pairing in *Drosophila*.

Finally, although triple pairing has been observed in the triploid chicken this does not prove that it exists in other triploids or that quadruple pairing exists in tetraploids. It does, however, suggest a need for a similar examination of other polyploid organisms.

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Persistence of Dose Related Behaviour in Mice

If naive rats are injected with an amphetamine-barbiturate mixture similar to 'Drinamyl' and are tested in an unfamiliar environment—a Y-shaped runway—their spontaneous activity is about double that of controls given only saline¹. If, after testing, the rats are returned to their home cages and retested in the runway without drugs, about half of the stimulant effect of the drug mixture can still be detected as much as

3 months later^{2,3}. This is surprising, for significant amounts of the individual drugs or of any known metabolites are unlikely to be present in the animals' brains for longer than at most 48 h^{4,5}, though as far as we know information of this kind is not available for mixtures of the drugs. In any case, no such behavioural after-effects can be detected after one or a series of "passive" administrations of the drugs, whereby the rats are merely injected and replaced in the home cage without being tested³ (R. D. Porsolt, unpublished results and refs. 6 and 7). Only when drug administration is combined with an actual test experience do these long term after-effects seem to occur. In the conditions described they are consistent and reliable and are presumably due to some more or less permanent changes in the organism, which are induced by the first drug/environment experience and which affect subsequent behaviour.

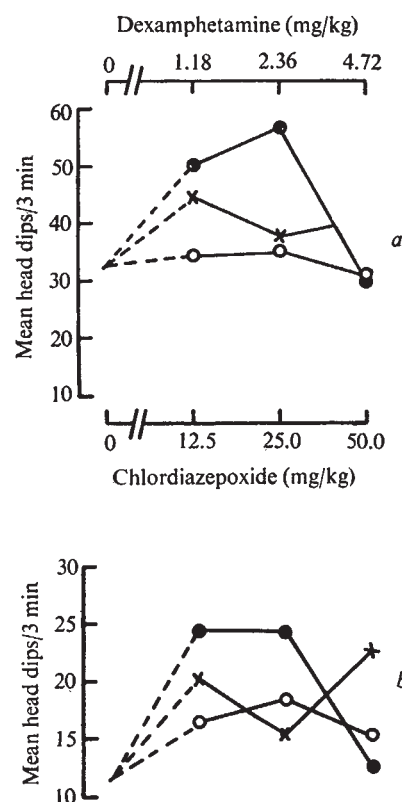


Fig. 1 Head dipping by mice under the influence of three doses of dexamphetamine (x) or chlordiazepoxide (O) given alone and in combination (●) in a constant ratio of approximately 1:10 by weight at trial 1 (a). The mice were tested on a hole board 20 min after intraperitoneal injections, and the number of times they dipped their heads into holes during a 3 min trial is shown on the ordinate. Each point represents the mean for eight mice and there were twenty-four controls. With one dose of the mixture, the amount of head dipping was greatly increased but the separate drugs had little effect. At trial 2 (b) 1 week later, curves of similar shape, though at a uniformly lower absolute level, emerged, even though the mice were now tested without any drug or injections. The standard errors ranged from 2.39 (saline) to 4.48 at trial 1 and 1.64 (saline) to 4.97 at trial 2. The scale of the ordinate for trial 2 has been doubled for easier comparisons of shapes.

To analyse long term effects of single drug experiences further, we have now used several doses of a different drug mixture, a different species of rodent—the mouse—and a different test environment—a hole board instead of a Y-maze. The drug mixture consisted of dexamphetamine combined with chlordiazepoxide ('Librium'—a drug which, like amylobarbitone, is widely used to allay anxiety) instead of amylobarbitone, for previous experiments with rats had shown

mixtures of these two drugs to be even more effective in stimulating activity than were amphetamine-barbiturate mixtures⁹. We used mixtures of drugs because they induce much greater increases in activity than any dose of the separate ingredient drugs⁹, and they therefore provide a favourable baseline from which to study after-effects.

Groups of eight naive female adult mice, Porton strain, and housed four per cage, were injected intraperitoneally with one of three doses of a dexamphetamine-chlordiazepoxide mixture in a ratio of 1:10 by weight, or with the separate constituents, or with saline. The four mice in a cage all received the same treatment. The particular ratio of the two drugs was chosen because previous Y-maze experiments with rats⁹ and hole board experiments with mice¹⁰, using amphetamine-barbiturate mixtures, had shown that dose ratios which were optimal in rats were also effective in mice. Twenty minutes after injection the mice were placed singly on square horizontal wooden boards with sixteen holes evenly spaced in four rows¹¹. Behaviour was scored during 3 min as follows: (1) total number of head dips: a dip was counted whenever the mouse put its head into a hole far enough for both its eyes to disappear below the surface of the board—to explore holes is very natural for mice; and (2) amount of walking about on the board, determined by drawing on specially prepared paper grids the path which the mouse followed on the board: the number of times the animal's path crossed the grid lines was taken as a quantitative index of the amount of walking. Other forms of behaviour such as grooming, defaecation and urination were also recorded.

After the original 3 min test (trial 1) the mice were replaced in their home cages and left for 1 week; they were then given their second test (trial 2) on the same boards but without any drug or other injections. Otherwise the procedure was identical with the procedure followed at trial 1.

The results show that at the first trial the separate drugs in any of the three doses used hardly affected the number of times that the mice dipped their heads into the holes, as compared with saline controls (Fig. 1, trial 1); with chlordiazepoxide most of the head dips were, however, characteristically concentrated in the first minute of the 3 min trial⁸. With the two smaller doses of the mixture, on the other hand, head dipping increased markedly as compared with saline ($P < 0.05$ and $P < 0.01$ for the lowest and for the middle dose of the mixture respectively, Mann-Whitney U test). With the biggest mixture dose, the amount of head dipping was no different from that of the controls, but the mice were very ataxic. When all the mice were retested, without any drugs, one week later (Fig. 1, trial 2) the shapes of the original dose-response curves seemed to re-emerge (Kruskal-Wallis one way analysis of variance, $H = 17.675$, d.f. 3, $P < 0.001$) though the absolute scores for all groups were approximately 50% lower than at trial 1. Fig. 1 shows results for all possible kinds of head dip combined; these include first dips into a hole, which accounted for the largest proportion of dips, repeat dips into the same hole, which occurred fairly often with amphetamine alone and with the 2.36:25 mixture dose, and dips over the edge of the board, which were rarer. The increases in repeat dips are consistent with the often reported finding that some doses of amphetamine are apt to produce various repetitive or "stereotype" movements. When the three kinds of head dip were plotted separately, a similar re-emergence of dose-response curves from trials 1 and 2 was found. As for the amount of walking on the board, again the separate drugs seemed to have little effect (Fig. 2, trial 1); the mixture, however, increased walking, especially in the smallest dose used ($P < 0.05$); with the highest dose, the amount of walking was no different from the saline controls, possibly because of ataxia. At trial 2, without drugs, the shapes of the curves again well resembled those of the dose-response curves at trial 1 (Kruskal-Wallis analysis of variance for the mixtures $H = 7.860$, d.f. 3, $P < 0.05$) except that mice which had received the biggest dose of dexamphetamine at trial 1 walked as much as naive controls,

which may have been due to an intensified amphetamine effect because of grouping. In all other cases the absolute levels at trial 2 were approximately half the trial 1 levels, as was found with the number of head dips. In general the variances in the walking results were larger than in the head dip results. The two kinds of measure were not significantly correlated, but they seemed equally sensitive to the drugs and to their after-effects.

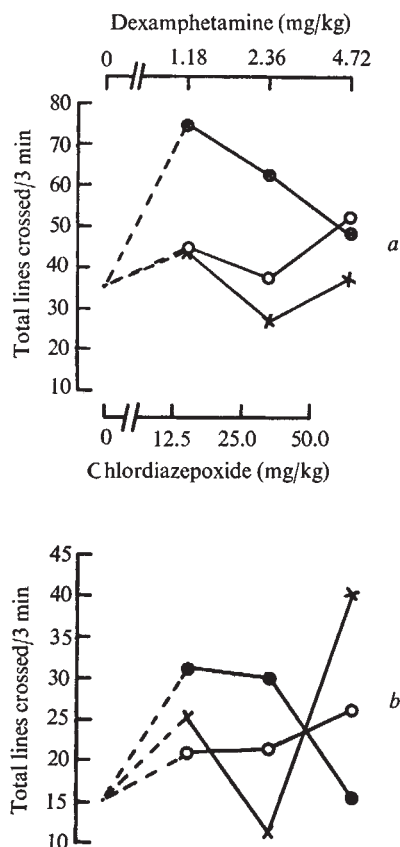


Fig. 2 Amount of walking over the hole board by mice treated as described in Fig. 1. Moderate doses of the drug mixture at trial 1 increased walking but the separate drugs did not have much effect. At trial 2, without drugs, except for mice which had had 4.72 mg/kg of dexamphetamine, the dose-response curves were recreated. The scale of the ordinate for trial 2 has been doubled for easier comparisons. Symbols are as in Fig. 1.

The problem of how much, if anything, of what has been learnt under the influence of drugs can be elicited later in the undrugged state has been much discussed. Various experimenters using drugs similar to ours have reported less retention the greater the original dose, and some have argued that with sufficiently large doses virtually nothing is learnt, and animals behave at the second, non-drug trial as though the first trial had not occurred; this phenomenon is often referred to as "dissociation" or "state-dependent" learning^{2,10,12-15}.

Our experiments show that not all drugs act in this way; with the highest dose the animals should have behaved more like naive controls than with the smaller. In fact, all animals seem to have reproduced a constant proportion of their behaviour, regardless of the original dose. The 50% reduction in activity at trial 2 with saline is what usually happens when mice are exposed for a second time to this kind of unfamiliar environment and may be regarded as a sign of reduced curiosity, or habituation. It seems that the mice which had been given drugs, similarly, "remembered" their drug-induced abnormal behaviour, and to some extent reproduced it, when the opportunity was given; from gross observation it also appeared that

at trial 2 the animals were more hesitant, sniffed more and walked less deliberately.

These experiments show that if the circumstances in which drugs are first encountered are appropriate, clear and dose-dependent effects on behaviour can persist for a surprisingly long time. A possible explanation could be that memory of the previous behaviour is reactivated by exposure to the original test situation.

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Identification of a Volatile Constituent formed by Homogenates of *Acacia georginae* exposed to Fluoride

We reported¹ that some homogenates of *Acacia georginae* and of other plants can convert added inorganic fluoride to a volatile compound, which is lost after alkaline combustion. We found examples in which the total fluoride estimated by acid diffusion after combustion was less than the fluoride estimated by acid diffusion without combustion. The first hypothesis which we examined was that vinyl fluoride ($\text{CH}_2=\text{CH}_2\text{F}$) was formed. Some evidence (although not decisive) was obtained for this by gas chromatography using a column of 'Poropak S'. We then explored the possibility that a volatile fluoroketone might be formed. Monofluoroacetone has been found in the

livers of rats perfused with fluoroacetate², and we record here evidence which suggests its presence as one of the volatile fluorine-containing constituents of the *A. georginae* homogenates.

Where possible, AnalaR reagents were used. Fluoroacetone was supplied by Aldrich Chemical Co. and AnalaR 2,4-dinitrophenyl hydrazine was recrystallized. Inorganic fluoride (and any fluoride freed by acid) and organically combined F^- were estimated by Hall's method³.

The gases from the homogenate, prepared and reinforced as described previously¹ during 1 h of incubation at 30° C, were drawn through a solution of 2,4-dinitrophenyl hydrazine (0.1% in 2 N HCl). The mixed hydrazones were extracted into ethyl acetate and treated as in the protocols.

For gas chromatography, compounds were prepared by drawing the gases from the homogenate through a tube (30 cm $\times$ 2.5 cm) containing CaCl_2 for approximately 2 h. An evacuated flask was then attached to the CaCl_2 tube, and gently warmed, so that the collected gases entered the flask, which was cooled in $\text{C}_2\text{H}_5\text{OH}/\text{CO}_2$. Controls showed that 20 μg amounts of both monofluoroacetone and acetone could be detected on the polyethylene glycol 600 column at 82° C after this treatment.

The "hydrazones" were spotted on to Whatman No. 52 paper, previously dipped in 30% phenoxyethanol/acetone, and blotted. The chromatogram was run for 48 h, in an atmosphere saturated with ligroin (boiling point, 80–100° C), the liquid phase being phenoxyethanol saturated with ligroin².

Controls showed that fluoroacetone can be recovered as 2,4-dinitrophenyl hydrazone from combustion mixtures and from plant homogenates. In our first experiment a homogenate, made by grinding in a mortar 2.7 g of leaves from a young plant of *Acacia georginae*, was strained through muslin to make a total volume of 7 ml. The plant had been watered weekly for more than 40 weeks with 200 ml. of a solution containing 200 μg of NaF. The homogenate was divided into two halves, each reinforced with adenosine triphosphate, pyruvate and phosphate¹, making a total volume of 3.6 ml., pH 7.4. Inorganic NaF, 50 $\mu\text{g}/\text{g}$, was added to one half. During incubation for 1 h at 30° C air was passed over the homogenates into an acid solution of 2,4-dinitrophenyl hydrazine (α). After the incubation, the hydrazones present were shaken six times with approximately 1/5 volume of ethyl acetate. A portion of the combined ethyl acetate extracts was taken to a low volume *in vacuo*, treated with LiOH and Mg succinate and ashed at 400° C. The fluoride in this fraction (α) was estimated by acid diffusion³; β , the residual aqueous extract from α , was neutralized and evaporated to dryness; the gases produced were drawn through acid 2,4-dinitrophenyl hydrazine and, using the same procedure as in α , fraction β was obtained.

Table 1 Fluoride Analyses on the 2,4-Dinitrophenyl Hydrazones obtained from the Vapours arising from Homogenates of *Acacia georginae*—Additions of NaF

Fraction	nmol F^-	
	No addition	+ NaF
α	58	630
β	157	157

For details, see the description of our first experiment.

Table 1 shows that the amount of a volatile ketone, behaving as fluoroacetone, is much increased by the addition of F^- to the homogenate. A control containing everything except the homogenate showed no volatile F^- .

In our second experiment a similar treatment was given to another homogenate from *A. georginae*. After extracting the "hydrazones" with ethyl acetate and their reduction to a suitable volume, they were run on paper chromatograms, as

described. Spots were found running in the same position as the 2,4-dinitrophenyl hydrazone of fluoroacetone, although acetone ran differently. As stated by the Japanese workers, acetaldehyde hydrazone ran in the same position as fluoroacetone hydrazone, but we failed to separate the two hydrazones of acetaldehyde and fluoroacetone on the column, as recommended². The presence of fluoroacetaldehyde could not be ruled out, although it seems improbable in view of its instability⁴.

This experiment shows that a volatile fluoro-ketone is present in the vapours above the homogenates. Further evidence was sought by gas chromatography.

In our third experiment, using the technique described, the vapours from a homogenate of *A. georginae*, treated with LiOH and Mg, were passed through the CaCl₂ tube while being gradually evaporated to dryness. After they had been transferred to the evacuated flask, the vapours were passed through the gas chromatograph and showed a peak corresponding in position to that of monofluoroacetone.

We have thus proved chemically that F⁻ leaves the homogenates of *A. georginae* in a gaseous form, and that some can be isolated as a 2,4-dinitrophenyl hydrazone, behaving at least in part as monofluoroacetone. The same fluoroacetone has been found in the vapours of homogenates treated as for combustion. It has therefore been proved that the apparent loss of F⁻ in our previous experiments¹ is attributable to the escape of fluoride in a volatile form. At the same time, calculation shows that the F⁻ collected in this experiment does not represent more than 13% of the total amount of F⁻ disappearing, so that there are probably other volatile F⁻ compounds still to be identified. It is interesting to note that fluoroacetone is relatively non-toxic, and that it could arise from fluoroacetic acid⁵.

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Visual Contour Masking

WE wish to report an effect we noticed accidentally while observing the parallel grid figure of Gregory's book *Eye and Brain*¹ (Fig. 1). When the page was stationary only the vertical grid would be seen; but during rapid to and fro, left to right movement the fins of the Müller-Lyer figure became visible. The latter happened to be on the reverse side of the page (136). Holding the page so that the grid was horizontal did not make possible the observation of the Müller-Lyer figure either when stationary or with left to right movement—although it could be seen during up and down movement.

This effect was confirmed on a group of twenty-two naive university students. The page with the parallel grid was removed from the book and affixed to a white card, so that the legend to the figure was not visible. The subjects were told that there was a figure behind the bars, and that they had to describe,

draw or guess what it was. Initially, the grid was horizontal and stationary, then it was moved to and fro from left to right. The procedure was repeated with the bars vertical and stationary, and then moving from left to right. Only in this last condition did the subjects report the presence of lines inclined to the grid, and drew the fins of the Müller-Lyer figure; sometimes the vertical stems were added or the outer ends were joined.

A similar effect can be observed with a ray figure of the type described by Mackay². If a circle is drawn on the back of a sheet of paper of suitable thickness with a ray figure on the front it cannot be seen when the ray pattern is stationary, but it is visible during rotation. It is unlikely that the complementary images produced by the patterning prevent perception of the circle, as blacking out a large central circle abolishes the complementary images but does not result in the appearance of the circle (when the pattern is stationary).

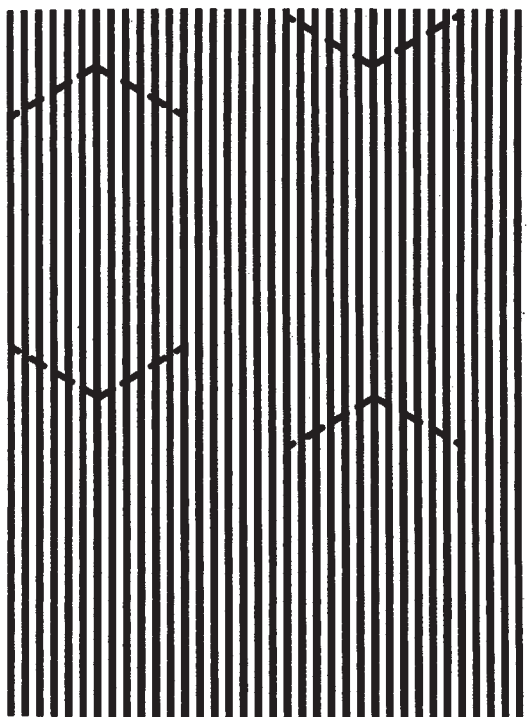


Fig. 1 The grid figure from Gregory's *Eye and Brain* (135). On the reverse page to this grid was a Müller-Lyer figure (in the position indicated by the broken lines which we have added). The Müller-Lyer figure could be seen during left to right movement, but not during up and down movements of the page.

These results are pertinent to recent work concerned with visual contour masking. A figure presented for some time masks another figure subsequently presented, if it is in or near the same orientation³. The point of interest in this communication is that the masking occurs simultaneously; that is, a figure with repetitive contours of high contrast can mask a figure of very low contrast inclined relative to it. Successive visual masking has proved amenable to neurophysiological interpretation in terms of orientation detectors^{4,5}, and the same approach can be applied to this effect. In this case the orientation-specific analysers stimulated by the parallel grid would inhibit the firing of analysers for other orientations (probably in a manner similar to that in binocular contour rivalry). During left to right movement of the vertical grid acuity would be reduced⁶. Presumably, this is accompanied by reduced stimulation of the orientation-specific analysers, with consequent reduction in the inhibition of other orientation analysers; it is in these conditions that the inclined figures are seen. When the grid is horizontal and moved from left to right, acuity is not

appreciably reduced and the analysers would therefore be stimulated in the same manner as the stationary pattern, with similar inhibition of inclined orientations. Lateral inhibition of orientation-specific analysers has been suggested to account for certain other visual effects⁷, and similar processes may underline the masking effects reported.

Simultaneous contour masking is evident in binocular rivalry and apparently also in normal perception. It is clear in this case that the contrast of the grid and the velocity of the figure are of prime importance in eliciting this effect, and these variables are at present under investigation.

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Induction of Pseudopregnancy in Rats by *p*-Chlorophenylalanine

p-CHLOROPHENYLALANINE (PCPA), a depletor of brain serotonin¹, has been reported to affect various categories of behaviour, including several patterns of explicitly conditioned responses²⁻⁶, as well as "spontaneous" activities, such as loco-

results to demonstrate that pseudopregnancy in adult rats can be induced by PCPA.

Adult rats of the Charles River CD strain were caged in groups of five, in a room which was lighted only from 0600 h to 1800 h. Food and water were always available. Vaginal smears were taken daily, and rats were used only after they had displayed at least two consecutive 4 day oestrous cycles immediately before the experimental cycle. PCPA was suspended in a steroid-suspending vehicle (SSV, National Institutes of Health) and injected as follows: 150 mg/kg, intraperitoneally, on the morning of oestrus of the experimental cycle (day 0) and a similar injection on each of the next 3 mornings. Control rats were injected with similar volumes of SSV. On day 4—that is, the next day of oestrus in the case of uninterrupted cyclicity—rats were either autopsied for the counting of tubal ova or subjected to traumatization of one uterine horn to test for the ability to produce a decidual response. Exceptions to these procedures are noted below. Rats undergoing uterine traumatization were autopsied on day 9. The results are discussed in two categories: (a) findings until day 4 and (b) findings after day 4.

Of twenty-three PCPA-treated rats, twenty had dioestrous vaginal smears on days 3 and 4; on these days all of sixteen controls displayed the sequence pro-oestrus-oestrus, as expected for normal cyclicity. This difference between PCPA-treated and control groups is highly significant ($P \leq 0.0001$, by the exact probability method). The three atypical PCPA-treated rats continued to cycle, although irregularly, and were not used further in the experiments. All of six control rats autopsied on day 4 had a full complement of fresh tubal ova (12.5 ± 0.9 per rat); none of six PCPA-treated rats killed on day 4 had tubal ova. This difference is significant ($P = 0.0011$).

Fourteen PCPA-treated rats, dioestrous on day 4, were subjected to uterine traumatization; five of these died during ether anaesthesia. Of the nine surviving, seven continued to have dioestrous smears until day 9, the day of autopsy. Six of these seven also displayed definitive decidual responses. The increment in uterine horn weight caused by traumatization was conspicuously greater for the PCPA-treated rats than for the controls (Table 1). (The difference is significant, $P < 0.002$,

Table 1 Body, Uterine and Ovarian Weights of PCPA-treated Pseudopregnant Rats and Controls

	Body weight (g)	Weight of traumatized uterine horn (mg)	Weight of untraumatized uterine horn (mg)	Weight of traumatized horn minus wt. of untraumatized horn (mg)	Weight of single ovary (mg)
SSV-treated controls (<i>n</i> =9)	275.9 ± 9.1	195.4 ± 13.6	169.6 ± 13.4	25.8 ± 1.5	45.1 ± 2.3
PCPA-treated pseudopregnant (<i>n</i> =6)	277.5 ± 9.6	1,478.3 ± 269.5	148.1 ± 15.2	1,330.2 ± 261.1	34.6 ± 1.8

Each entry is a mean ± s.e.

motion⁷ and social interaction⁸, and particularly sexual behaviour. Tagliamonte *et al.*⁹ demonstrated that PCPA increased homosexual activity in male rats, and while confirming this¹⁰, they showed that the effect was potentiated by testosterone. Sheard¹¹ reported that heterosexual behaviour of rats was stimulated by PCPA, but Whalen and Luttge¹² could not confirm this result. In ovariectomized rats, PCPA was found to be a substitute for progesterone in the restoration of oestrous behaviour¹³. In male cats, PCPA was reported¹⁴ to promote homosexual activity; but this report has been challenged¹⁵.

In spite of considerable interest in the effects of PCPA on sexual behaviour, there is little information about its possible effects on reproductive physiology, except a brief report¹⁶ that it delayed the onset of vaginal opening, and of cyclicity in the vaginal smear of young rats. We now have preliminary

by the two-sample rank test.) Thus, by day 9, six of the nine rats were still pseudopregnant, as judged by both decidual sensitivity and vaginal smears. One of the other three rats still had a dioestrous smear, but its decidual response was doubtful. In the remaining two, the pseudopregnancies indicated by the smear on day 4 had clearly been interrupted by day 9. The results for SSV-treated controls clearly contrast with those for PCPA-treated rats. Ten controls were subjected to uterine traumatization; one died under ether. All nine surviving controls continued to show cyclicity in their vaginal smears. Eight maintained 4 day cyclicity, and one had a 5 day cycle. None of the nine had any decidual response. The difference between PCPA-treated and control groups in the incidence of pseudopregnancy on day 9 (six out of nine against nought out of nine) is significant ($P = 0.0045$).

A further difference between the PCPA-treated, pseudo-pregnant rats and the controls is that the former showed reduced ovarian weights (Table 1). This difference is significant ($P < 0.01$) and does not seem to be a result of differences in body weights, which do not differ significantly ($P \gg 0.10$).

Although we demonstrate here the induction of pseudo-pregnancy by PCPA, our work does not indicate the mode of action of the drug in causing this effect. Previous work suggests that depletion of brain serotonin may be involved¹. But it should be noted that at least one study¹⁷ has indicated that concentrations of brain noradrenaline, as well as serotonin, are reduced by PCPA. If this is so, the action of PCPA may be similar to that of reserpine, which has been shown to cause pseudopregnancy in rats¹⁸ and to deplete both brain serotonin¹⁹ and brain noradrenaline²⁰. The comparison of reserpine with other drugs which fail to cause pseudopregnancy has suggested that depletion of brain noradrenaline is the cause of the effectiveness of reserpine²¹. Further research will be necessary to determine which aspect of the activity of PCPA causes pseudopregnancy. It is possible that this peripheral effect of PCPA is independent of changes in the biochemistry of the central nervous system.

The high mortality rate of PCPA-treated rats during anaesthesia, although not statistically significant, suggests that PCPA may alter sensitivity to anaesthesia.

Our results demonstrate the desirability of determining the basic reproductive physiological effects of PCPA. Such information may resolve the controversies already noted concerning the effects of PCPA on sexual behaviour.

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Red Photoreceptor in Butterflies

It has recently been suggested¹ that modifications of the tracheoles beneath the photoreceptors of the compound eye of the butterfly *Heliconius erato* specifically reflect red light and that this increases the sensitivity of the eye to red light. Because *H. erato* has a red forewing patch which plays a role in the release of courtship activity, it was also proposed that the reflexions were related to the behavioural responsiveness of the animal to the colour red. We wish to comment on the behavioural and physiological implications of this observation.

From a behavioural standpoint, the responsiveness of *H. erato* to various colours is a labile characteristic². Although the butterflies respond to red in courtship, they have a spontaneous preference for yellow in feeding behaviour, for example. It is also known that the close relative, *H. charitonius*, can be conditioned to select virtually any colour in its feeding behaviour^{3,4}. Preliminary experiments (unpublished) suggest that *H. erato* is equally responsive to such conditioning and that such adaptability must indeed be of considerable importance in this polymorphic species⁵ which, in various localities, may have a forewing patch that is red, yellow, white or even entirely absent.

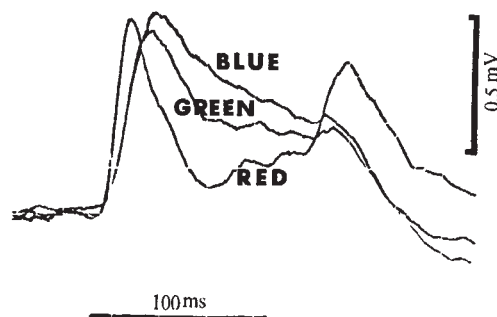


Fig. 1 Three superimposed electroretinograms recorded from *Heliconius erato*, with a subcorneal Pt electrode. Responses were produced by 100 ms flashes illuminating the whole eye. Near monochromatic stimuli were produced by narrow-band interference filters with maxima at 420, 528, and 621 nm. Intensities were adjusted to produce nearly equal amplitude initial negative deflexions. Note that the ERG produced by the red stimulus contains the largest positive component (downward deflexion following the initial negative peak) while the blue elicits the least.

It is also relevant that the same red "glow" seems to be a characteristic of at least the entire subfamily Heliconiinae. We have examined the eyes of four species, including the most primitive member of the group⁶, *Philaethria dido* (a green and brown butterfly); an intermediate representative, *Dryas julia* (orange and black); and two members of the genus *Heliconius*—*H. cydno* (black and white) and *H. sarae* (black and yellow). Regardless of wing colour, the eyes of all these butterflies have produced the same red glow when viewed with an ophthalmoscope. Even the iridescent blue *Morpho* butterflies respond strongly to red during courtship, their eyes reflecting red light in the same way.

We also wish to comment on the implication¹ that the spectral sensitivity of the butterfly eye is similar to that of a moth, with differences due solely to internal reflexions rather than differing arrays of photopigments. This conclusion was based on a consideration of the electroretinogram (ERG). We consider, however, that careful evaluation of such potentials suggests the presence of a photopigment with maximum absorption in the red part of the spectrum. The ERG of *H. erato* is a complex diphasic waveform, consisting of at least two components, one a cornea-negative effect considered to represent a summation of receptor potentials, and the second a cornea-positive com-

ponent originating deep in the retina. The spectral sensitivities of the two effects are markedly different, which means that it is impossible to match the ERG waveforms elicited by various wavelengths (Fig. 1). Differences in waveform are evident even in near-threshold responses; the positive component seems larger at longer wavelengths, and has a maximum at about 620 nm (Fig. 2) which corresponds to the "narrow peak" reported by Bernhard *et al.* These two responses are physiologically independent and can be reversibly isolated from each other by altering the inorganic ion balance. A high Na⁺ concentration, for example, favours the negative component, while at low Na⁺ concentrations the positive polarity effect is distinct. Clearly the physiologically different effects produced by stimulation with the red part of the spectrum cannot be explained in terms of simple reflexion.

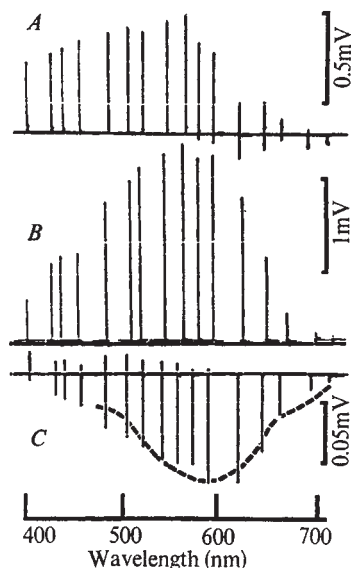


Fig. 2 Spectral efficiency plots recorded directly on an oscilloscope in response to 100 ms flashes of monochromatic light. The horizontal deflection of the beam was proportional to the wavelength of the stimulating light. Vertical deflexions indicate the maximum negative (upward) and positive potentials developed during stimulation (off responses were excluded from recording). A, Responses to sixteen interference filters, adjusted to deliver nearly equal energy (ca. 0.05 mW to the entire eye), recorded from a normal, intact individual, with a subcorneal Pt electrode. B, Recording as in A, except that the individual was treated with subcorneal Na⁺ so that it produced simple monophasic, negative polarity ERGs. C, Recording as in A, except that low Na⁺ saline was added to the head capsule so that the individual produced simple monophasic positive ERGs. Dashed line indicates theoretical absorption of a retinal based photopigment, calculated after the method of Dartnall¹⁰.

Electroretinograms and evoked potentials in the visual pathway of *H. erato*^{7,8} reveal several physiological phenomena with action spectra with peaks in the red, and with a shape more consistent with the absorption spectrum of a retinol based photopigment than with the narrow peak of an interference effect.

Recently, higher order single units in the visual pathway of the butterfly *Papilio troilus* have been reported⁹ which respond specifically to the stimulus wavelength. Such neurones are excited by narrow portions of the visible spectrum and are intensity independent; they demonstrate an opponent-process type mechanism. We have obtained similar results from a study of *H. erato*. Units were recorded which responded specifically to blue, orange, red, deep red and so on. Clearly, basal reflexions could not provide the nervous system with the information required to make such fine distinctions in stimulus wavelength.

It is axiomatic that colour vision is independent of intensity. Even if the basal reflexions could increase the absolute sensi-

tivity of the eye, they could neither enhance nor diminish the animal's ability to discriminate a given wavelength. In our opinion, the response to red colour is a function of the nervous system, not of the structure of the eye itself.

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Flying Ability of *Archaeopteryx*

HEPTONSTALL¹ has applied certain formulae given by Pennycuik² to demonstrate that the wing bones of *Archaeopteryx* were strong enough to support the bird in flapping flight. I have approached³ this problem by using some of Pennycuik's other work⁴. Basically, my estimates³ of the probable flying speed and also the wing loading for *Archaeopteryx* of approximately 7 m/s and 0.4 g/cm² respectively, depend, like the estimates of torsion derived by Heptonstall¹, on the estimation of the weight and wing area of *Archaeopteryx*.

Heptonstall, following Jerison⁵, supposed that *Archaeopteryx* weighed about 500 g. The wing span of *Archaeopteryx* was, however, only 58 cm, and arboreal birds of this size only weigh 200–250 g⁶. (Mammals of this body size also weigh about 200–250 g.) Only very heavily built birds, galliform or charadriiform genera such as *Lagopus*, *Scolopax* and *Phasianus*, weigh as much as 500 g. Jerison⁵ does not say how he obtained his weight estimate, but I feel sure it is at least twice too heavy.

Heptonstall's figure¹ for the wing area is 373 cm²; my estimate of this parameter is 479 cm², but my estimate of the area of the two wings alone is 388 cm², fairly close to the figure Heptonstall gives. I think he has overlooked the fact that "A" in his formula

$$v = \sqrt{\frac{2L}{C_L \rho A}}$$

includes, by accepted convention, the body strip between the wings. This adds another 91 cm² to the figure.

Without reworking Heptonstall's data, it is evident from these figures that his estimates of *Archaeopteryx* flying ability are pessimistic and likewise his comments on the potentially high landing speed of the bird. *Archaeopteryx* was arboreal, and had only to climb, losing flying speed as it did so, until it stalled on the tree canopy with very little forward speed.

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In a recent letter to *Nature*, Heptonstall¹ states that he used the same formula as Whitfield and I used to calculate the stalling speed of *Pteranodon*. I should like to point out that the formula, used with a high lift coefficient, gives the stalling speed, or minimum possible flying speed, not the maximum velocity as he states.

The body weight of *Archaeopteryx* has recently been recalculated³ as 200 g, a value far below the 500 g obtained by Jerison⁴ and used by Heptonstall. This new calculation seems to me to be far more reasonable and its use alters the wing loading, the stress on the bones and the flying characteristics of the animal. The wing area given of 373 cm² is too low; the true figure is 479 cm². This includes a piece of body between the wings, in accordance with standard aeronautical convention; this is done by Pennycuik⁵ and others⁶ working on bird flight.

But the most serious false assumption in this article is the statement that the lift generated by the large tail of *Archaeopteryx* can be ignored because it is too far back to matter in normal flight. This tail had an area of 140 cm² and so approached the dimension of one of the wings. Although the tail is behind the centre of gravity of the animal, the lift produced is still significant, particularly when the centre of lift of the wings moves forwards during the downstroke of the wing beat. Incidentally, in the Concorde, much of the lifting surface is at the rear and in conventional planes the horizontal tail flaps provide an additional lifting surface placed well to the back of the aircraft. The tail of *Archaeopteryx* was also important in a variety of other ways; in providing stability, for example.

I have recently been trying to determine the aerodynamic characteristics of *Archaeopteryx* so as to test the animal's gliding performance, using a computer program designed to do this; this has already been done for *Pteranodon*⁷. One of the most difficult problems has been finding the lift and drag coefficients for the tail. Already it can be estimated that the lift from the tail reduces the stalling speed by approximately 20%. This is a useful adaptation for low speed landing and makes it a less violent business than that envisaged by Heptonstall. It certainly would make things easier if the tail of *Archaeopteryx* could be ignored, but it is a large integral part of the animal and must be considered in any sensible aerodynamic analysis.

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DR HEPTONSTALL writes: Both Yalden¹ and Bramwell² question the weight of *Archaeopteryx* which I estimated to have been 500 g (ref. 3). My figure was based on a detailed anatomical comparison with the pigeon, *Columba livia*, the average weight

of which is nearly 400 g (ref. 4). Although the lengths of many of the bones are similar (for example, femur, radius, ulna and metacarpals) some are larger (for example, fibula and humerus). The comparison showed that certain parts of the body were significantly heavier, particularly the head, neck and tail. The weight of the organs in the thorax of *Archaeopteryx* can only be inferred from an estimate of the volume. Using Heilmann's reconstruction⁵, I found that the volume for *Archaeopteryx* was very similar to that of the pigeon. From this I deduced that *Archaeopteryx* could not have weighed less than 400 g (that is, at least twice that suggested by Yalden), and after allowing for the additional weight of the head, neck, tail and skeleton (the bones are not pneumatic) I arrived at a figure close to 500 g. In merely comparing the wing span with various living species, Yalden misses the point that this primitive bird had a high wing loading (probably higher than that of the pheasant⁶).

The omission of the body strip in my calculations is implied in my sentence which begins "If the body . . . provided negligible lift". This strip was only 40 cm² (not 91 cm² as implied by Yalden's data) and its inclusion leads to a figure for lift which is too high. It is often more convenient to use it but it is a convention which is not strictly valid since the body strip does not behave like an aerofoil in generating high lift. I considered that in view of the possible errors already incurred by selecting typical values for C_L and k , the effect of the body strip could be ignored. Although some authors incorporate this strip, others do not^{4,7}.

I agree with Yalden's comment concerning flight paths from tree to tree. I was making the point that the bird seems to have been poorly adapted for landing on the ground.

Bramwell's first statement concerning the use of the classical equation in aerodynamics relating lift force to velocity and the like is incorrect. Keeping the values of C_L , ρ and A constant, the general relationship then follows that the lift force is proportional to the square of the velocity and after defining the maximum permitted value of L the equation obviously provides maximum v . In her criticism of the omission of the tail from the calculations, Bramwell claims that because the tail is big it must generate appreciable lift. This is clearly not true because any flat (or nearly flat) surface must be inclined several degrees to the flow before lift can be generated⁸. I consider that while *Archaeopteryx* glided, just sufficient lift was induced to balance the weight of the tail. This elaborate tail undoubtedly had its uses, particularly for stabilizing or inducing pitching movements, and may well have been used to a limited extent in flapping flight as Bramwell envisages. The main function of the tail plane of an aircraft is to provide stability like the feathers on an arrow, not for giving it lift.

In conclusion, I should like to point out that Yalden's opening sentence is misleading in that the formula which I used relating bone strength to bending moment is that which is widely quoted in textbooks on strength of materials and was not used by Pennycuik in his work on pigeon flight⁴.

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- ¹ Yalden, D. W., *Nature*, **231**, 127 (1971).
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³ Heptonstall, W. B., *Nature*, **228**, 185 (1970).
⁴ Pennycuik, C., *J. Exp. Biol.*, **46**, 219 (1967).
⁵ Heilmann, G., *The Origin of Birds* (Appleton, New York, 1927).
⁶ Romer, A. S., *Vertebrate Palaeontology* (University of Chicago, Chicago, 1966).
⁷ Alexander, R. McN., *Animal Mechanics* (Sidgwick and Jackson, London, 1968).
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BOOK REVIEWS

Echoes of Science

The Royal Institution Library of Science: Physical Sciences. Edited by William Lawrence Bragg and George Porter. (Being the Friday Evening Discourses in Physical Sciences held at the Royal Institution 1851–1939.) Vol. 1: Pp. 577. Vol. 2: Pp. 549. Vol. 3: Pp. 569. Vol. 4: Pp. 548. Vol. 5: Pp. 569. Vol. 6: Pp. 450. Vol. 7: Pp. 511. Vol. 8: Pp. 536. Vol. 9: Pp. 550. Vol. 10: Pp. 477. (Elsevier: Barking, Essex and Amsterdam, 1970.) £63.00.

THE Royal Institution *Library of Science* is a straight reprint of the Friday Evening Discourses from 1851 to 1939, arranged by subject in a consecutive series; the ten volumes on physical science were first issued some months ago and are now completed by an index volume which makes their use much more convenient. Two volumes on astronomy have also been published (see *Nature*, 228, 951; 1970) and other series are planned. The discourses are in full or in abstract, as originally published; thus Vol. 2 has the whole of Tyndall's splendid essay "Faraday as a Discoverer" (74 pp.) while in Vol. 1 a lecture by Faraday himself on ozone ends with the flat précis: "The evening concluded with the expression of certain theoretical expectations, or rather possibilities, which were put forth as indicating the probable fertility and importance of the subject..." Since the reprint is a photographic one there is no danger of textual errors, only mechanical ones (such as have unfortunately but not misleadingly occurred in Vol. 5, where I also note the error of "Some Recent Results of Physico-Chemical Injury" (Inquiry) in the contents page).

Such a massive reprint is, of course, the raw material for history, not history itself. Any worthwhile commentary or annotation for a work of this scale would be an enormous task, and (probably wisely) none has been attempted. If (again in Vol. 5) the reader is puzzled by J. J. Thomson's use of Lenard's table of results on the penetration of cathode rays—where there seems incidentally to be an original misprint of 0.760 mm for 760 mm—in his celebrated lecture of

April 30, 1897, concluding with Thomson's determination of the e/m ratio—he must seek elucidation elsewhere; similarly he must establish for himself the cross-connexion with Thomson's later discourse of 1901 in the same volume. Again, he must observe for himself that Charles Brooke's date (Vol. 1, p. 24) for the introduction of the compound microscope is egregiously incorrect. These are the minutiae to which the reader of original historical documents must always be alert. Far more interesting is the *Library of Science's* presentation of the progress of the physical sciences actually witnessed by the members of the Royal Institution, which was not always in accord with the chief lines as we commonly see them today. Vol. 1 provides an excellent conspectus of mid-Victorian science, with Faraday (past his greatest years, but always lucid and original), Tyndall, Grove and Odling as its leading exponents; electromagnetism, the unity of forces, organic chemistry are, as one might expect, dominant themes. H. E. Roscoe gave (1861) a clear account of the significance of the recent development of spectroscopy, while Helmholtz delivered a superb lecture in the same year on the conservation of force. As the reader moves forward, noting the appearance of the synthetic dyestuffs industry and the solitary discourse by Clerk Maxwell ("Action at a Distance", 1873) he enters the long continued age of Crookes, Dewar, William Thomson and Rayleigh, with Mendeléef (1889) offering the rather rare contribution of a distinguished foreigner (his lecture adumbrated chemical dynamics—it is perhaps worth noting in this same context Vernon Harcourt's 1868 anticipation of the law of mass action). Because it is not, perhaps, surprising to find A. W. Hofmann postulating a "light metallic" ammonium radical (NH_4) nor recurrent insistence on the reality of the aether, it is with the last decade of the nineteenth century that the pattern of discourses seems to vary from the pattern of modern historical writing. True, Vol. 4 (1890–1896) takes in the work of Hertz and Tesla, the discovery of argon, the problem of aether-drift, and early work on low-temperature electromagnetism.

But it does not seem that the Michelson–Morley experiment exerted a decisive influence on thought (it was described by Lodge in 1892 and by Kelvin in 1900, both discounting its result for different reasons). X-rays were the subject of two discourses, in 1896 and 1899, the latter containing a useful theoretical review; however (apart from an allusion to radium by J. J. Thomson in 1901), radioactivity remained undiscussed until Becquerel himself was invited to give a discourse as late as 1902, an invitation likewise extended to Pierre Curie in the following year and to Rutherford in 1908. Moreover, apart from the discovery of the inert gases, very little attention was given to chemistry by the Royal Institution during the last decade of the nineteenth century. One cannot but gain the feeling that, with a few distinguished exceptions, the chief foci of new ideas and experiment in physical science at this time lay outside Britain.

With the first two decades of the new century, however, one of the most fertile epochs in the history of British experimental physics, the discourses are dominated by the contributions of J. J. Thomson and his brilliant pupils. If the Royal Institution heard the word quantum for the first time, apparently, only in 1914 (W. H. Bragg), and if such elder statesmen of science as Dewar and Rayleigh appeared frequently, radioactivity, the positive rays, cloud-chamber photography, atomic disintegration and its radiant energy, the alpha particle, all were described at Albemarle Street by most distinguished exponents—among them another French visitor, Jean Perrin (on Brownian movement, 1911), X-ray crystallography was first discussed by W. H. Bragg in 1914, and by his son in 1920; after the elder physicist's appointment as director of the Royal Institution in 1923 it was the subject of annual reports. At this point—in the last two volumes of the set—the names of living scientists appear; besides Sir Lawrence Bragg, Professor Andrade, Sir Harold Hartley, Lord Blackett, Sir Eric Rideal, Sir George Thomson and many others no longer living whose names are still equally familiar. Rutherford gave no fewer than nine discourses in the years

1924-1939. In fact Vol. 10 ushers in the nuclear age and, with Professor J. D. Bernal's discourse on "The Structure of Proteins", the age of molecular biology also.

"The crucial fact that requires elucidation," Bernal declared in 1939, "is the precise mode of folding or coiling of the peptide chains, and for this we may have to wait for some considerable time until the X-ray and other methods have advanced their technique much further than at present. The problem of the protein structure is now a definite and not unattainable goal, but for success it requires a degree of collaboration between research workers which has not yet been reached. . . . It is difficult to exaggerate the importance of this study to many branches of science."

The editors and publishers are to be congratulated on the republication of so much important material in an accessible, if hardly cheap, series of volumes. If it must be remembered that a Friday Evening Discourse is rarely the first announcement of a new achievement in science, nor often descriptive of its early stages, it is almost invariably an authoritative review, and one that has the advantage of allowing the lecturer to explain and sometimes to speculate more freely than in a formal scientific paper. Thus it may convey a rich enlightenment of its own. One plea, finally, I would add—that the publishers will print an alphabetical table of all the contributors represented in the *Library of Science*, with the titles of their discourses, since at the moment one must search each volume separately.

A. RUPERT HALL

Maltese Prehistory

The Prehistoric Antiquities of the Maltese Islands: a Survey. By J. D. Evans. Pp. ix + 260 + 70 plates. (Athlone: London, April 1971. Distributed by Tiptree: Tiptree, Essex.) £15.00.

THE great stone temples of Malta are among the most remarkable monuments of the prehistoric west Mediterranean, and indeed of Europe in general. Their grandeur, the great skill displayed in the transport of the huge stones and in their dressing, and the sophistication of the decorative art which they contain, are without parallel in the Old World beyond the lands of the early civilizations.

They present, then, one of the enigmas of history, and up to twenty years ago little could be attested with confidence of their date or origins. Systematic clearance work and excavation by the Maltese archaeologist Sir Themistocles Zammit had yielded quantities of material, but opinions and dates proposed by scholars ranged very widely. This

monumental work by the professor of prehistoric archaeology at the London Institute of Archaeology, the fruit of a decade's labour, shows how radically the previous rather confused picture has been transformed.

In 1952, the author was invited to catalogue the extensive material in the National Museum of Antiquities in Malta, much of it from Zammit's excavations, and to conduct a survey of prehistoric sites. The most important product of this study was the division of Maltese prehistory, on the basis of the preserved finds, into a sequence of eight prehistoric cultural periods, as set out by Evans in the *Proceedings of the Prehistoric Society* (19, 41; 1953). This gave, for the first time, a coherent chronological framework into which the major developments of Maltese prehistory could be fitted. The construction of the temples themselves was assigned to the Ġgantija and Tarxien phases, named after the two principal monuments of Gozo and Malta respectively.

The catalogue of prehistoric sites, with the finds from them, forms the bulk of the present volume and was prepared during the period from 1952 to 1958. The author had the collaboration of a number of workers, listed in the preface, in the preparation of plans, drawings, photographs and a full bibliography of Maltese prehistory. The plans are particularly welcome, documenting an entire and much neglected chapter in European architectural development.

The production of this volume has been a lengthy process, and despite the very high quality of its presentation by the Athlone Press, I must express both surprise and regret that four years elapsed between the completion of the manuscript and its eventual publication. The long gestation of the work has, however, happily allowed the inclusion of the findings of Dr David Trump, who was curator of antiquities in the National Museum from 1958 to 1963. His excavations at the Skorba temple confirmed stratigraphically the sequence established by Evans, and extended it with the addition of three new phases (as well as the repositioning of another). Trump's work yielded valuable new material, and furnished an important series of organic samples for radiocarbon dating. Evans has accepted Trump's modifications, so that the sequence here presented is now universally accepted.

This handsome volume thus contains virtually all that we know of the prehistory of Malta, arranged now in a coherent and stratigraphically documented culture sequence and supplemented by radiocarbon dates. It is one of those rare and authoritative works which at once becomes a classic, and which a century after publication will not have been superseded.

Prehistoric Antiquities of the Maltese

Islands is, of course, a corpus. It is not a book for reading but for reference. Of the 260 pages of text, only the introduction (5 pages) and the last chapter, "Culture and Chronology" (22 pages), make easy and continuous reading. The bulk of the book is given over to a description of each of the known sites and of the artefacts there found. As well as the famous four great temples (Ġgantija, Tarxien, Mnajdra, Haġar Qim) and the four or five more well known European prehistorians, Evans lists and discusses a further 15 sites with megalithic (large stone) remains, as well as caves, tombs, settlements and dolmens of the later prehistoric period. The finds are described and illustrated by findspot rather than by period, so that no coherent picture of Maltese prehistory is to be gained by leafing through the illustrations. For this one must turn rather to Evans's *Malta* (Thames and Hudson; 1959), a book now regrettably out of print.

When the value of massively documented presentations of archaeological material has sometimes come into question, it is important to assert that this amply illustrated, comprehensive treatment is the very stuff of archaeology. No serious discussion of prehistoric Malta can ever again be conducted without reference to the evidence *in toto*, as here so conveniently assembled. Indeed, the level of discussion is automatically raised to a new plane. Evans has wisely separated his data from the interpretation given in the final chapter, although this is itself both careful and cautious. He discusses Trump's conclusion, based on the radiocarbon dates, that the Tarxien phase (and with it the period of the later temples) ended about 2000 bc: "The present author, however, finds himself unable to share this view, feeling that the evidence for contacts with Crete in the later Middle Minoan period, and perhaps with the beginning of Mycenaean culture, is too strong to be denied . . . the most likely date for the beginning of the Tarxien phase seems to me to be some time after 2000 bc, and it ended not before 1600 bc" (p. 223). Since Evans wrote these words, the tree-ring calibration of radiocarbon has necessitated that ^{14}C dates be set much earlier, and I have suggested (*World Archaeology*, 2, 205; 1970) that the Tarxien phase be dated from about 2900 bc to about 2300 bc. This would have the remarkable consequence of making the earlier Maltese temples the earliest free-standing stone monuments in the world. More Maltese dates are needed to make this convincing, however, even if the calibration be accepted. My point here is that such a change in dating, while it would necessitate revision to the final chapter, would make little difference to the body of this volume. Changes in interpreta-

tion do not threaten the integrity of the corpus.

The corpus, on the other hand, will certainly modify interpretations. The very number of "temples" and megalithic structures reported must suggest new functions and meaning. Every village settlement must have had one, and the next step will be some sort of locational analysis to see if the four or five really large temples might plausibly be seen simply as the major building for a large village, or whether they had some "metropolitan" function. My own guess is that the term "temple" is too specialized in its implications, and that we should think rather in terms of communal centres—parish halls as much as parish churches. There are plenty of ethnographic parallels for buildings used by the society as a whole (or by a large part of it) for a wide range of activities, some of them decidedly secular. We do not have to imagine the prehistoric Maltese going about their daily affairs in "a state of worship", as some authors would wish.

This splendid volume will offer new insights or lines of approach to many archaeologists. For example, the presentation of the Tarxien Cemetery material suggests to me that far closer Aegean parallels for this post-temple period will be found (in Early Helladic III and Middle Helladic Greece) than for the earlier phases. The bronzework admittedly could be earlier, but the bossed bone plaque, the one and two-handled cups and the multiple vessels all find echoes in middle bronze age Greece as well as in the Capo Graziano culture of the Lipari islands.

The systematic presentation of this material, then, marks a real step forward in Maltese prehistory. It does not pretend to tell us why the temples were built or the sculptures carved, but it does describe them fully and systematically, document the material equipment of their builders, and lay the indispensable groundwork for determining just when they were built. Professor Evans has given us a work which does justice to the remarkable, indeed amazing, creations of which he writes.

COLIN RENFREW

Grand Mastery

Computers, Chess and Long-Range Planning. By M. M. Botvinnik. Translated by Arthur Brown. (Heidelberg Science Library, Vol. 11.) Pp. xiii + 89. (Longmans: London, February 1971.) £2.50.

BOTVINNIK's profound style of chess made him one of the greatest masters in the history of the game. For some time he has been researching into computer programs for playing chess, and his work has been awaited with considerable

interest. In this book he develops the principles which he believes should be the basis of a good chess program.

The book has two sections. In the first, Botvinnik gives his mathematical formulation of chess. The central idea is that of the intangible values of a piece; these are calculated in terms of the enemy men which the piece can attack in one or more moves. Botvinnik proceeds to give formulae for deciding whether a variation seems favourable, whether it should be examined further and so on. An interesting innovation is his "horizon method": in examining a possible move one considers only those pieces that can reach the relevant squares within a preassigned number of moves. Some remarks are offered on the difficult task of formalizing positional play. In the second section Botvinnik takes issue with the general practice of teaching the pieces how to make single moves. He gives instead tables ("clichés") which show the number of moves each piece requires to reach a given square on a clear board. This approach may well prove valuable.

It has been generally realized for some years that restrictions on the variations considered are necessary if the computer is to be capable of seeing deeply enough into the position. Relevant principles—development of pieces, control of the centre and so on—are familiar enough, but how are they to be programmed? At present no computers can play better than a moderate amateur. The real test of Botvinnik's ideas is whether they result in a significant improvement. Botvinnik, however, has not yet produced a workable program. He gives, as an "experiment", his celebrated combination against Capablanca, attempting to show that it could have been discovered by application of his theories. But he offers no reason why the computer should analyse the winning line of play to a depth of some twenty-five half moves (with Botvinnik a piece down throughout!). To find the line, the machine would need ideas about the value of advanced passed pawns, ideas not mentioned in the book. Nor is there any mention of the possibility of computers learning from experience, even though this seems likely to be the basis of future developments in the field.

The general outline of the argument is usually clear enough, but too often the technical detail is sketchy and the formalism obscure, to say the least. A typical example is the "important remark" on p. 16. The translation is competent and the presentation attractive, though there are a number of misprints. Unfortunately the price is quite unrealistic, for the text is only some fifty pages, supplemented by sixty positions "for study"—which seem perfectly useless—and some mildly relevant appendices. Also, the title has been altered, as part of an attempt to produce a coffee-table science book. The original title, "An Algorithm

for Chess", is more appropriate for what is essentially a blemished but interesting technical paper.

D. J. STRAUSS

Ion Movements in Cells

Membranes and Ion Transport. Vol. 2. Edited by E. Edward Bittar. Pp. xi + 295. (Wiley-Interscience: London and New York, December 1970.) £5.50.

THE publication of books on membrane function takes place with increasing frequency and there ought therefore to be some good reason for publishing a new, three volume analysis of the subject. The stated intention is to undertake a general survey of membrane function and to treat the subject both systematically and critically. The first volume, dealing chiefly with the organization of membranes and the more theoretical aspects of membrane transport, generally satisfies these two criteria. The second volume, which is concerned with ion movements in symmetrical cells and subcellular organelles, is less successful.

What I found disturbing is not so much that the survey lacks completeness—the omission of any reference to ion transport in plant cells surely not being justified—as that some authors show much more critical awareness of their subject than do others. This variability, which must of course always be present to some extent in a book of this nature, is highlighted in the present instance by the way the editor has chosen to present his subject. More or less equal space has been allowed in the first part of the book for articles on ion transport in seven different tissues. This is in spite of the fact that much more work needs to be reviewed by authors dealing with nerve, red blood cells and skeletal muscle than with any other tissue. The result has not been to cramp the reviews of ion transport in these three tissues—indeed, that by Hurlbut on nerve is excellent in all respects—but rather to encourage the remaining articles to expand out of proportion to their natural content. Part of this expansion takes the form of repetition. Is it necessary, for instance, to repeat potted definitions of transport and diffusion or to summarize the discovery of transport ATPase and describe the action of cardiac glycosides in quite so many parts of the same book, particularly when these subjects have been spoken about at length in the first volume of this series? Then must we have in duplicate the hoary old argument, which really has no answer, about whether it is better to use tissues *in vivo* or *in vitro* to study ion transport (ion movements in smooth muscle and brain)? Incidentally, the use of brain slices of 35 mm thickness (page 156) is not likely to advance the cause for *in vitro* work (the correct value is 0.35 mm). Further duplication arises in describing the use and misuse of extra-

cellular space markers. The objection is not that these points are unimportant but that they have been treated in an unsystematic fashion.

There is also a tendency to describe ion movements in cells rather than ion movements across cell membranes. No one would deny the importance of ion-binding in certain tissues in producing concentration gradients which are not all they seem. The description of such phenomena should obviously find a place in any book concerned with ion transport. What we get, however, is a description of calcium binding in relation to excitation-contraction coupling and relaxation (ion movements in heart muscle). Elegant as this description is, its relevance to membrane transport is minimal. Ion movements in mitochondria and across the nuclear envelope have been dealt with adequately. The importance of selective transport of ions across the nuclear envelope has still to be established, but what is known was presented in such a way as to encourage one to find out more about the processes involved.

There is much in this book of value to the animal physiologist who is prepared to sift through the occasional irrelevances. The sections on ion transport in nerve, skeletal muscle and red blood cells, as well as the articles on subcellular organelles, are well worth reading, but the student of plant physiology will find little of interest in this volume.

M. W. SMITH

Anatomical Information

New Research in Plant Anatomy. Edited by N. K. B. Robson, D. F. Cutler and M. Gregory. (Supplement to the Botanical Journal of the Linnean Society, Vol. 63.) Pp. xii+250. (Academic: London and New York, January 1971.) £6.00.

THIS collection of twenty papers is a tribute to Dr C. R. Metcalfe on the occasion of his retirement from the Keepership of the Jodrell Laboratory at Kew. It is also the first report of a Linnean Society Symposium to be published as a Supplement to the Botanical Journal of that Society.

In the introduction, Dr Metcalfe remarks on the contribution which studies of internal morphology using the light-microscope have made to the increase of knowledge in fields as varied as physiology, ecology, morphogenesis and systematics. As might be expected on this occasion, the majority of the papers illustrate the value of anatomical information in systematics and taxonomy. Five of them deal with relationships between species: Ayensu (*Dioscorea*), Brittan (*Thysanotus*), Kukkonen (*Carex*), Stace (*Juncus*), Stern (*Ribes*); and four of them with relationships between families: Cheadle (xylem of Liliales *sensu* Hutchin-

son), Clifford (numerical taxonomy of anatomical and other characters in Monocotyledones), Philipson (Umbellales), Stant (affinities of the monogeneric Petrosaviaceae). Another four papers are concerned with wood anatomy: Carlquist (xylem of subdendroid spp. of *Euphorbia*), Chalk (xylem fibres of *Fraxinus*), Findlay and Levy (scanning-beam electron-microscopy of wood) and Greguss (Araucariaceae). The remaining papers are of a more specialized nature. Those by Vaughan and by Chowdhury and Buth deal with the identification of seed fragments in feeding-stuffs, reminding us of the applied aspects of plant anatomy. Scott and Bystrom give some information on the mucilage cells of *Hibiscus* while the paper by Fahn and Rachmilevitz describes the ultrastructure of the nectariferous hairs of the hypanthium of *Lonicera*.

There is something in this collection for everyone who has an interest in plant anatomy or systematics. A few examples must suffice. Who can doubt the close relationship between the predominantly tropical and arborescent Araliaceae and the largely temperate and herbaceous plants taxonomists have designated as the Umbelliferae in the light of Philipson's contribution? Clifford's suggestion that grasses may have arisen from the palms by way of the bamboos particularly intrigued me. The same notion had occurred to me, when visiting the Palm House at Kew, on the evidence of *gestalt* coupled with Cornerian intuition. Evidence which confirms one's prejudices is always welcome!

Stace's caution against seeing inter-specific hybrids of *Juncus* in every wet meadow is admittedly controversial but his anatomical evidence accords with their reluctance to be hybridized in cultivation. On the other hand, I was startled by Greguss's suggestion (p. 85) that thin-walled marginal cells of the wood rays of *Agathis* produce more ray cells in the manner of a cambium. His plates do not convince me, and a rapid examination of prepared slides of the wood of two other species from the collection of the late Professor Jane serves to further increase my doubts. I am also inclined to disagree with Carlquist's premise (p. 182) that *Euphorbia* is a "basically herbaceous genus, several sections of which have become woody under various circumstances". The other genera of the family are predominantly woody and less specialized than *Euphorbia*. Consequently, it is probable that the subdendroid euphorbias which survive on island refuges or in xeric environments represent relatively archaic elements while the more numerous and widespread herbaceous species have been derived by neoteny.

The contributors to the symposium offer us some stimulating ideas as well as new information. The editors and pub-

lishers have also played their part by ensuring that the text and illustrations are well presented. The volume reflects the progressive attitude of the Linnean Society of London today and makes a fitting retirement present for the eminent botanist to whom it is dedicated.

B. M. G. JONES

Animal Behaviour

Comparative Animal Behavior. By Richard A. Maier and Barbara M. Maier. (Core Books in Psychology Series.) Pp. viii+459. (Brooks/Cole, a division of Wadsworth: 1970. Distributed in the UK by Prentice Hall International, Hemel Hempstead.) £5.50.

THIS latest addition to the long line of textbooks on animal behaviour brings together in a compact form some useful and interesting information from various areas of the subject. But there are a number of weaknesses in this book which would seem to render it unsuitable for teaching purposes at almost any level.

First of all, the book totally fails to mention a large number of major papers in areas that it specifically covers. For example, the first section is called "Structural and physiological foundations of behaviour" and we might reasonably expect this to include important work in physiology relevant to behaviour. Under the heading of "Vision", however, there is no mention of Hubel and Wiesel's work on the physiology of the mammalian visual system; surely knowledge of this work is essential to any student of behaviour. In a section entitled "The Role of Sensory Cues in Migration and Navigation" (p. 255) there is no reference to G. Matthew's theory of Sun navigation by birds, and only a brief mention of the fact that the Sun may be important in bird orientation at all. In a discussion of the effect of early deprivation on sensory processes (p. 321), Held and Hein's work on the development of visuo-motor coordination is not included. These three examples are not obscure, nor are they simply illustrations of points made with reference to other material. They are serious omissions from a text that claims to be comprehensive, and they are by no means the only ones.

This apparent neglect of the literature often leads to statements being made on the basis of old work which we now know to be incorrect or wrongly interpreted. On p. 325 we read that "When rats are raised in environments without access to anything that can be manipulated, e.g. straw, paper, or even their own faeces, the results are more extreme: these animals fail to build nests at all (Riess, 1950, 1954)". As long ago as 1963, Eibl-Eibesfeldt showed that this failure of Riess's rats to build nests was probably due to their being tested in a strange cage,

where even normally reared rats will explore rather than build a nest. When rats deprived in this way are tested in their home cage, they build perfectly good nests. This point has been reiterated in almost all recent animal behaviour textbooks.

Just as serious as what is left out of this book are some of the things which are put into it. There are a number of generalizations which are based on so little fact that they amount to virtual falsehoods. For example, it is stated on p. 219: "The structure of a school is generally regular, with approximately the same number of fishes in each of its three dimensions". In fact, we know very little about the structure of fish schools in any species. And it is misleading to enumerate the functions of play in young animals as if they had been demonstrated with certainty (p. 225).

The basis of the distinction between "higher" and "lower" animals, which is made throughout the book, is not explained (the octopus is described as a "lower invertebrate") and the title of Part 2, "Functional Behaviour Patterns", carries the somewhat curious implication that some behaviour patterns are non-functional.

These criticisms may sound unnecessarily severe. They have been expressed strongly because there are now so many books about animal behaviour that the publication of yet another seems justified only if it meets a high standard and has some particular contribution to make. On both these counts this book is unfortunately somewhat lacking.

MARIAN DAWKINS

Measure for Measures

Hausdorff Measures. By C. A. Rogers. Pp. viii + 179. (Cambridge University: London, December 1970.) £3.80: \$12.50.

THE book is divided into three chapters, and the titles are respectively "Measure in Abstract, Topological and Metric Spaces", "Hausdorff Measures" and "Applications of Hausdorff Measures".

The first chapter is on general measure theory and contains the fundamentals of the subject. It would be suitable for somebody learning measure theory for the first time. Although the treatment is abstract it is not unduly general. There is considerable emphasis on measures in metric spaces, for this theory is needed in the second chapter.

The second chapter is undoubtedly the chief one. Although the material follows quite logically from the first chapter, the non-specialist might find it difficult. Nevertheless, it is very readable, and a particularly useful feature is that outlines of proofs of theorems are sometimes given before the details. Much of the existing literature of the subject is

collected together, and some of the topics are generalized and developed further. Most of the chapter is concerned with general metric spaces, but the case of Euclidean space is briefly discussed in relation to Lebesgue measure and surface areas. One of the principal topics concerns existence theorems. For example, various conditions on the space are proved sufficient to ensure that a compact set of infinite Hausdorff measure contains a compact set of positive finite measure. In this connexion there is a reference to a joint paper by Roy O. Davies and the author in which an example is given to show that some restrictions on the space are needed. It seems a pity that this interesting example is not included in the book.

The third chapter is concerned chiefly with Euclidean space and is therefore less abstract. The title of the chapter is somewhat misleading as many of the topics are not so much applications of Hausdorff measure as part of geometric measure theory studied in its own right. Most of these topics are only briefly surveyed. The survey is reasonably complete but there are some omissions. The two topics considered in detail are continued fractions and continuous increasing functions.

This is not just another book on measure theory and it is unique in the ground covered. It is remarkably self-contained and there is an excellent bibliography. It should be particularly useful to the beginning research mathematician who is interested in measure theory and to the specialist as a source of reference.

J. M. MARSTRAND

Structure and Reaction

Chemical and Mechanical Behaviour of Inorganic Materials. By Alan W. Searcy, David V. Ragone and Umberto Colombo. (Accademia Nazionale dei Lincei, Eleventh Course of the Guido Donegani Foundation, Tremezzo (Lake Como), Italy, September 8–20, 1968.) Pp. xxiv + 715. (Wiley-Interscience: New York and London, December 1970.) £13.00.

THIS large book contains the lectures presented to a summer school arranged "for graduates in chemistry and physics who have minimal backgrounds in materials science", people who are assumed to know little of crystal defects or of solid-state chemical reactions. To the materials scientist's jaded palate it is a refreshing, because unusual, diet; 29 chapters deal first with general chemical and defect principles, and subsequently with particular applications of some of these principles. The early chapters lean heavily on thermodynamic interpretations: thus one of Searcy's five chapters illuminatingly interprets the thermo-

dynamics of reactions involving solids and gases largely in terms of the entropy of gaseous products, and this is developed in de Maria's chapter on unfamiliar vapour molecules. The statistical mechanics of evaporation/condensation equilibria and the stability of various refractories are analysed in two complementary chapters.

The book moves on to structural aspects of solids. Heavy emphasis is placed in two chapters by Ragone on the physical chemistry of defects in solids and on their relation to the extent of non-stoichiometry. A particularly illuminating chapter later in the book is that by Reiss on the "electron as a chemical entity in solids". In effect, the electron is treated as a defect in equilibrium with donors and acceptors, and the origin of solute/solute solubility cross-effects in semiconductors is very clearly set out on mass-action principles. Reiss's chapter, which is frankly didactic in a field where many authors strive only to be encyclopaedic, is of a special quality: one is reminded of Hinshelwood's style in his *Structure of Physical Chemistry*.

Other chapters—more familiar to a materials scientist—deal with diffusion (Ascoli) and, very comprehensively, with dislocation geometry and dynamics and the special behaviour of dislocations in controlling ductility and fracture in ceramics (Conrad). Kelly surveys fibre-composites. Various aspects of surfaces are considered in several chapters. Ragone is interesting on topics such as the distinction between surface tension and energy, but somewhat short on practical implications of surface energy, such as thermal etching. Chemical surface (gas/solid) reactions also receive due attention.

The last part of the book deals with particular classes of materials. Zarzycki elegantly surveys the basic features that characterize the structure of oxide glasses, Bokros surveys the world of pyrolytic carbon which is almost his own creation, and two chapters deal with the very special problems of nuclear fuels. Another chapter surveys a number of special materials such as glass-ceramics and ferroelectrics (really too short to be very useful), and another provides extensive insight into ceramic-based batteries. This chapter is scientifically and technologically particularly interesting. The rear is brought up by Frank, who writes on the Earth as a ceramic body. He writes with all his familiar verve and éclat; it is most instructive to observe how many qualitative deductions about the nature of heat transport and tectonic processes in the Earth's core, mantle and crust can be drawn with the help of cunning order-of-magnitude calculations. A book much concerned with the man-made thus finishes with a deep look at levels inaccessible to man.

R. W. CAHN

CORRESPONDENCE

Lightning Balls

SIR,—I know of two instances of fireballs (the Yorkshire term for ball lightning). (1) A fireball came out of a fire-grate and flashed across the room to enter a radio, immediately putting it out of action. On later examination the valves were found to be damaged. (2) The second instance happened in the same street but was seen from outdoors. A fireball was seen to enter the chimney of a house. I do not know what was seen in the house on this occasion but electrical equipment was again found to be damaged after the incident.

As electrical equipment was damaged both times it is most unlikely that there was optical illusion, as suggested by Argyle¹. In any case, Argyle's mathematics are wrong. If the number of persons reporting ball lightning observations is 44% of the number reporting observation of ordinary lightning impact, this cannot be understood by assuming that about half of all strokes to ground

generate a luminosity ball at impact. If half the strokes to ground generate a ball, the number of ball reports would be about the same as that of ordinary lightning reports. In its simplest terms, if A equals B , then A is 100% of B . Not 44%.

Perhaps we should look at eyewitness accounts with less scepticism and more thought. We are told: (1) that ball lightning enters buildings and aircraft—it does not simply strike; (2) that fireballs may bounce several times on hitting the ground; (3) that they damage electrical equipment.

Fork lightning discharges to ground on impact. It would seem that ball lightning cannot discharge so easily, hence its bouncing behaviour on hitting level ground; hence also its tendency to earth through electrical equipment. The stable electrical configuration of most matter may cause it to be less attractive to ball lightning than is the "electron stream" offered by working electrical equipment.

Another cause for this apparent diffi-

culty in discharging might be assumed from the shape of ball lightning. Its "ballness" could indicate a relative stability due to some centre-point of attraction within the ball itself. I am not qualified to comment on whether such nuclear attractant might consist of matter or anti-matter², but I think it highly probable that it exists. It is also possible that what ends its journey as ball lightning may have begun its journey as fork lightning, which found and coalesced around material particles on the way down.

Yours faithfully,

MARGARET LAZARIDES

*Yeolmbridge House,
Launceston,
Cornwall*

¹ Argyle, E., *Nature*, 230, 179 (1971).

² Ashby, D. E. T. F., and Whitehead, C., *Nature*, 230, 180 (1971).

Announcements

University News

Professor D. N. S. Kerr has been appointed to the chair of medicine and headship of the Department of Medicine in the University of Newcastle upon Tyne.

Dr I. J. Maddox has been appointed to the chair of pure mathematics and Dr M. P. Haggard to the chair of psychology, in the Queen's University of Belfast. The title of honorary professor has been conferred on Dr G. F. Adams (geriatrics), Dr M. G. Nelson (haematology) and Dr J. Frank Partridge (cardiology).

Appointments

Dr T. L. V. Ulbricht, formerly managing director and director of research of Wyford Laboratories Ltd, has been appointed head of the newly formed Planning Section of the Agricultural Research Council, which will be concerned with the formulation of forward policy.

Dr J. D. Hood, a member of the MRC external scientific staff, has been appointed director of the Hearing and Balance Unit, recently established by the Medical Research Council.

Miscellaneous

The following have been elected fellows of the Australian Academy of Science: Professor G. Burnstock, University of

Melbourne; Dr W. Compston, Australian National University; Dr L. T. Evans, CSIRO Division of Plant Industry; Professor F. Gibson, Australian National University; Dr A. K. Head, CSIRO Division of Tribophysics; Professor G. A. Horridge, Australian National University; Professor L. E. Lyons, University of Queensland; Professor R. L. Martin, University of Melbourne; Professor G. E. Wall, University of Sydney; Dr D. E. Weiss, CSIRO Division of Applied Chemistry.

The new Chemical Society awards, each sponsored by an industrial company, have now been announced. The award for industrial chemistry, sponsored by Imperial Chemical Industries, has been made to Professor R. C. Pitkethly; for kinetics and mechanism, sponsored by Shell Research, to Professor J. C. Polanyi; for main group element chemistry, sponsored by Albright and Wilson, to Professor M. F. Lappert; for natural product chemistry, sponsored by Roche Products, to Professor D. H. R. Barton; for organometallic chemistry, sponsored by Monsanto Chemicals, to Professor J. Chatt; for structural chemistry, sponsored by the British Petroleum Company, to Professor A. Carrington; for theoretical chemistry and spectroscopy, sponsored by Varian Associates, to Professor A. D. Buckingham. Nominations for the 1971 awards, which will be made in the fields of chemical analysis and instrumentation, macromolecules and poly-

mers, medicinal chemistry, surface and colloid chemistry, synthetic organic chemistry, thermodynamics and electrochemistry and transition metal chemistry, are now invited. Further details can be obtained from Dr John F. Gibson, The Chemical Society, Burlington House, London W.1V 0BN.

International Meetings

June 25, **Current Topics in Cancer Therapy**, Paris (Professor E. H. Cooper, Department of Experimental Pathology and Cancer Research, University of Leeds, Leeds 2).

July 5-9, **Animal Behaviour**, Durban (Mr D. R. Masson, CSIR, PO Box 1, Congella, Natal).

July 14-16, **Modern Industrial Inorganic Chemistry**, Kingston upon Thames (Dr I. L. Stephenson, School of Chemical Science and Technology, Kingston Polytechnic, Penrhyn Road, Kingston upon Thames, Surrey).

July 25-30, **International Congress of Pure and Applied Chemistry**, Boston (Secretariat, c/o American Chemical Society, 1155 Sixteenth Street, NW, Washington DC 20036, USA).

July 26, **Mechanism and Regulation of DNA Replication**, Aspen, Colorado (Dr A. R. Kolber, Division of Infectious Diseases, Washington University School of Medicine, Saint Louis, Missouri 63110, USA).

August 26–29, **Pollution Control**, Lund (SSRS Office, 221 Rock Hill Road, Bala-Cynwyd, Pennsylvania 19004, USA).
 August 29–September 5, **Soil Water Physics and Technology**, Rehovot (The Organizing Committee, POB 16271, Tel Aviv, Israel).

September 6–8, **Pneumatic Transport of Solids in Pipes**, Cambridge (H. S. Stephens, Pneumotransport I, British Hydromechanics Research Association, Cranfield, Bedford).

September 6–9, **Physics and Chemistry of Asbestos Minerals**, Louvain (Mr B. Lincoln, Turner and Newall Asbestos Fibre Laboratory, c/o TAC Construction Materials Ltd, PO Box 22, Trafford Park, Manchester M17 1RU).

September 7–16, **Mycological Congress**, Exeter (Professor J. Webster, Hatherly Laboratories, Prince of Wales Road, Exeter EX4 4PS, Devon).

September 8–9, **High Voltage Insulation in Vacuum**, London (Meetings Officer, Institute of Physics, 47 Belgrave Square, London SW1).

September 8–10, **Electron Mean-Free Paths in Metals**, Brighton (Meetings Officer, The Institute of Physics, 47 Belgrave Square, London SW1).

September 13–16, **Liquid Scintillation Counting**, Brighton (Mr M. A. Crook, Society for Analytical Chemistry, 9–10 Savile Row, London W1X 1AF).

September 14, **The Use of Coordination Complexes in Organic Synthesis**, Manchester (Dr R. Fields, University of Manchester Institute of Science and Technology, PO Box 88, Sackville Street, Manchester M60 1QD).

September 14–16, **Solid State Devices**, Lancaster (Meetings Officer, Institute of Physics, 47 Belgrave Square, London SW1).

September 14–17, **European Biophysics Congress**, Baden, near Vienna (Intercongress, A-1010 Vienna, Stadiogasse 6, Austria).

September 16–17, **Explosion Hazards in the Chemical Industry**, Manchester (Dr B. J. Tyler, Department of Chemistry, UMIST, Sackville Street, Manchester M60 1QD).

British Diary

Monday, May 17

Celebrations to Mark the Centenary of the Foundation of the Institution of Electrical Engineers (three days, London).

Constructivity and Quantization (5.30 p.m.) Professor C. Kilmister, British Society for the Philosophy of Science, in the Joint Staff Common Room, University College London, Gower Street, London WC1.

Tumour Viruses and Cell Growth (5 p.m.) Dr M. G. P. Stoker, University of London, at St Mary's Hospital Medical School, Wright-Fleming Institute, Norfolk Place, London W2.

Tuesday, May 18

Changing Attitudes to Analytical Control of Medicinal Substances (4.15 p.m.) Mr C. A. Johnson, Society for Analytical Chemistry, jointly with the Loughborough University of Technology Chemical Society, at Loughborough.

Faraday and the Beginnings of Useful Electricity (5.30 p.m.) Professor Ronald King, Royal Institution, at 21 Albemarle Street, London W1. (Lecture for Fourth Form Pupils from Schools in London and the Home Counties. To be repeated on May 19, 25 and 26.)

National Libraries' Kaleidoscope (5.30 p.m.) Miss Maysie Webb, Royal Institution, Library Circle, at 21 Albemarle Street, London W1.

Rehabilitation of the Cancer Disabled (9.15 a.m. symposium) Marie Curie Memorial Foundation, in the Edward Lumley Hall, Royal College of Surgeons of England, Lincoln's Inn Fields, London WC2.

The Experimental Approach in Chemical Pathology (5.15 p.m.) Professor D. M. Matthews, University of London, in the Meyerstein Lecture Theatre, Westminster Medical School, 17 Horseferry Road, London SW1.

Wednesday, May 19

Factors in Anaesthetic Deaths (5.30 p.m.) Professor J. A. Thornton, British Postgraduate Medical Federation, University of London, at the Institute of Dental Surgery, Eastman Dental Hospital, Gray's Inn Road, London WC1.

London Discussion Meeting (6.30 p.m.) Society for Analytical Chemistry, Microchemical Methods Group, at Imperial College, London SW7.

Sporulation in Bacteria as a Simple Model for Differentiation in Higher Organisms (2 p.m.) Professor J. Mandelstam, University of London, at the Royal Postgraduate Medical School, Hammersmith Hospital, Du Cane Road, London W12.

The Changing Pattern of Medical Service in Tropical Africa (6 p.m.) Professor P. G. Janssens, Royal Society of Tropical Medicine and Hygiene, jointly with the Société Belge de Médecine Tropicale and the Nederlandse Vereniging voor Tropische Geneeskunde, at the Royal College of Physicians, 11 St Andrew's Place, Regent's Park, London NW1.

The Permissive Society (5.15 p.m.) Professor O. R. McGregor, University of London, at Bedford College, Regent's Park, London NW1.

Trevithick's Place in Engineering History, Mr James Hodge; **Richard Trevithick, William Smith and Samuel Homfray—Their Correspondence on Steam Engines 1804–1806**, Mrs J. M. Eyles, Institution of Mechanical Engineers, jointly with the Newcomen Society, at 1 Birdcage Walk, London SW1.

Thursday, May 20

515th Meeting (two days) Biochemical Society, at the University College of North Wales, Bangor.

Diseases of Fish (two-day symposium) Zoological Society of London, in association with the Fisheries Society of the British Isles, in the Meeting Room of the Zoological Society of London, Regent's Park, London NW1.

Health as a Factor for Socio-Economic Advance of Developing Countries (10 a.m. all day meeting) Royal Society of Tropical Medicine and Hygiene, jointly with the Société Belge de Médecine Tropicale and the Nederlandse Vereniging voor Tropische Geneeskunde, at the Royal Society of Tropical Medicine and Hygiene, 26 Portland Place, London W1.

Prelude to Power; A Light in Nature (1 p.m. films) Royal Institution, at 21 Albemarle Street, London W1.

Relationship Between Structure and Function of Antibody-Producing Cells (5.30 p.m.) Dr B. Pernis, University of London, at the London School of Hygiene and Tropical Medicine, Keppel Street, Gower Street, London WC1.

Vertebrate and Insect Haemoglobin (5.30 p.m.) Professor G. Braunitzer, University of London, in the Anatomy Theatre, University College London, Gower Street, London WC1.

Friday, May 21

Equatorial Currents in Atmospheres and Oceans (11 a.m. symposium) Royal Astronomical Society, jointly with the Royal Meteorological Society and the Challenger Society, in the Scientific Societies Lecture Theatre, 23 Savile Row, London W1.

Metal-Ion Photosensitized Reactions (1 p.m.) Dr R. J. Kemp, Royal Institution, Photochemistry Discussion Group, at 21 Albemarle Street, London W1.

Metals with Memory—Ferro-Mechanical Behaviour and Phase Transitions (9 p.m.) Professor David S. Lieberman, Royal Institution, at 21 Albemarle Street, London W1.

The Scientific Examination of Paintings and Antiques (7 p.m. conversazione) Dr A. E. Werner, Society of Chemical Industry, Fine Chemical Group, at 14 Belgrave Square, London SW1.

Monday, May 24

Power Switching by Vacuum Contactors and Thyristors (5.30 p.m. discussion) Institution of Electrical Engineers, at Savoy Place, London WC2.

The Teaching of Electrical Circuit Theory (5.30 p.m. discussion) Institution of Electrical Engineers, at Savoy Place, London WC2.

Reports and Publications

not included in the monthly Books Supplement

Other Countries

Annual Report of the Rubber Research Institute of Malaya for 1969. Pp. 143. (Kuala Lumpur: Rubber Research Institute of Malaya, 1970.) [123]

US Department of Commerce—National Oceanic and Atmospheric Administration. NOS Publication 31-3: Surface Water Temperature and Density—Pacific Coast, North and South America and Pacific Ocean Islands. Third edition. Pp. 88. (Washington, DC: Government Printing Office, 1970.) \$1. [123]

1970 Annual Report of the International Nickel Company of Canada, Limited. Pp. 32. (Copper Cliff, Ontario: International Nickel Company of Canada, Ltd., 1970.) [123]

US Department of the Interior: Geological Survey Professional Paper 546: The Alaska Earthquake, 27th March, 1964—Lessons and Conclusions. By Edwin B. Eckel. Pp. vi+37. (Washington, DC: Government Printing Office, 1970.) \$0.75. [153]

Australia: Commonwealth Scientific and Industrial Research Organization. Land Research Series No. 28: Lands of the Ord-Victoria Area, W.A. and N.T. Comprising papers by G. A. Stewart, R. A. Perry, S. J. Paterson, D. M. Traves, R. O. Slatyer, P. R. Dunn, F. J. Jones and J. R. Sleeman. Pp. 134. (Melbourne: CSIRO, 1970.) [153]

1970 Annual Report of the Southwest Research Institute. Pp. 44. (San Antonio, Texas: Southwest Research Institute, 1971.) [153]

US Department of Commerce: National Bureau of Standards. Selected Tables of Atomic Spectra A: Atomic Energy Levels—Second Edition. B: Multiplet Tables. CI, CII, CIII, CIV, CV, CVI. Data Derived from the Analyses of Optical Spectra. By Charlotte E. Moore. (NSRDS-NBS 3/3.) (Washington, DC: Government Printing Office, 1970.) \$1. [153]

Institut Royal des Sciences Naturelles de Belgique. Mémoire No. 163: Recherches sur les Eaux Saumâtres des Environs de Lilloo. Quelques Considérations Ethologiques à Propos des Travaux de W. Conrad sur les eaux Saumâtres de Lilloo. Par H. Kufferath. Pp. 39. Mémoires. Deuxième Série, Fasc. 84: Les Bivalvia Fossiles du Cénozoïque Étranger des Collections de l'Institut Royal des Sciences Naturelles de Belgique. VU (fin): Oligodontina (2), Astartodontina et Septibranchida. Par Maxime Glibert and Luv Van de Poel. Pp. 185. Mission Zoologique Belge aux Iles Galapagos et en Eciador (N. et J. Leleup, 1964-1965). Résultats Scientifiques, Deuxième Partie. Pp. 235. (Bruxelles: Institut Royal des Sciences Naturelles de Belgique, 1970.) [153]

Smithsonian Contributions to the Earth Sciences, No. 2: Catalog of Chemical Analyses of Rocks from the Intersection of the African, Gulf of Aden, and Red Sea Rift Systems. By Paul A. Mohr. \$2.75. Smithsonian Contributions to Zoology, No. 80: Freshwater Triclad (Turbellaria) of North America. IV: The Polyparyngeal Species of Phagocata. By Roman Kenk. Pp. 17. \$0.30. A Snetzler Chamber Organ of 1761. By John T. Fesperman. Pp. 56. \$0.70. (Washington, DC: Smithsonian Institution Press, 1970.) For sale by US Government Printing Office. [153]

Comptes Rendus des Travaux du Laboratoire Carlsberg. Vol. 38, No. 1: The Degradation on the B-Chain of Oxidized Insulin by Mucor Miehiei Protease. By William Rickert. Pp. 1-18. Vol. 38, No. 2: Pinocytic Uptake of Ferritin by the Amoeba *Chaos chaos* Measured by Atomic Absorption of Iron. By C. Chapman-Andersen and S. Christensen. Pp. 19-58 + 1 plate. Vol. 38, No. 3: Hydrogen Isotope Exchange of Insulin—a Comparison of Methods. By G. Ronca and L. Willumsen. Pp. 59-66. Vol. 38, No. 4: Studies on the Opal Apparatus of *Tetrahymena pyriformis* GL. By A. Forer, J. R. Nilsson and E. Zeuthen. Pp. 67-86 + 8 plates. (Copenhagen: Danish Science Press, 1970.) [153]

US Department of the Interior: Geological Survey. Bulletin 1294-H: The *Foerstia* Zone of the Ohio and Chattanooga Shales. By J. M. Schopf and J. F. Schwietering. Pp. iii+15+2 plates. \$0.30. Bulletin 1267: Bibliography of North American Geology, 1967. Pp. xxx+1029. \$4.25. Water-Supply Paper 1964: Quality of Surface Waters of the United States, 1965. Parts 7 and 8: Lower Mississippi River Basin and Western Gulf of Mexico Basins. Pp. xiii+819. \$3.50. (Washington, DC: Government Printing Office, 1970.) [163]

US Department of Commerce: National Bureau of Standards. NBS Special Publication 319: Man, His Job, and the Environment—a Review and Annotated Bibliography of Selected Recent Research on Human Performance. By William G. Mather and Boris V. Kit. Pp. vi+101. \$1. NBS Special Publication 305: Publications of the National Bureau of Standards 1968-1969. By Betty L. Oberholtzer. Pp. xxii+497. \$4.50. (Washington, DC: Government Printing Office, 1970.) [163]

The Biology of *Atriplex*. Edited by R. Jones. (Studies of the Australian Arid Zone. Based on a Symposium held in Deniliquin, NSW, 14/15th October, 1969. Pp. 129. (Canberra: Division of Plant Industry, CSIRO, 1970.) [163]

CSIRO, Australia. Annual Report of the Wool Research Laboratories for 1969/1970. Pp. 70. (Melbourne: CSIRO, 1970.) [163]

Pharmaceutical Manufacturers Association, Washington. Annual Survey Report, 1969/1970. Pp. iv+20. (Washington, DC: Pharmaceutical Manufacturers Association, 1970.) [163]

Texas Instruments, Inc., 1970 Annual Report. Pp. 8. (Dallas: Texas Instruments, Inc., 1971.) [163]

CSIRO, Australia. Annual Report of the Division of Tribophysics, 1969/1970. Pp. 22. (Melbourne: CSIRO, 1970.) [163]

Harvard University Programme on Technology and Society. Sixth Annual Report 1969/1970. Pp. 103. (Cambridge, Mass.: Harvard University Program on Technology and Society, 1970.) [163]

Annals of the New York Academy of Sciences. Vol. 171, Article 1: International Conference on Singlet Molecular Oxygen and Its Role in Environmental Sciences. By R. W. Murray *et al.* Pp. 1-302. \$25.50. Vol. 171, Article 3: Metabolism and Biological Functions of Polyamines. By E. J. Herbst *et al.* Pp. 691-1009. \$26. (New York: New York Academy of Sciences, 1970.) [173]

Fiskeriundersøgelser i 1969 ved Danmark. Færøerne og Grønland. Ved E. Bertelsen og Paul M. Hansen. Pp. 101. (København: I kommission hos Andr. Fr. Høst and Son, 1970.) [173]

Space and the United States Navy. By Commander Ted Wilbur. Prepared by the Editors of *Naval Aviation News*. Pp. 80. (Washington, DC: Government Printing Office, 1970.) \$1.75. [173]

Preservice Preparation of College Biology Teachers. A Search for a Better Way. By Donald S. Dean. Pp. x+122. (Washington, DC: The Commission of Undergraduate Education in the Biological Sciences, 1970.) *Gratis*. [173]

Metallographical Interpretation and Evaluation of Defects in Winding Rope Wires. By Sri R. P. Chakraborty and Sri T. K. Sengupta. Pp. 38. (Dhanbad, Bihar: Central Mining Research Station, 1968.) Rs. 2. [173]

Science Council of Canada. Canada, Science and the Oceans. (Report No. 10, 1970.) Pp. 37. (Ottawa: Science Council of Canada, 1970.) [173]

Katalog Polskiej Literatury Biologicznej/Catalogue of Polish Biological Literature, Tom II (XXIII) za Lata 1955/1959. Czesc 2: Zoologia, Paleontologia, Rozne. Pp. xiv+479. (Krakow: Academie Polonaise des Sciences, 1970.) [173]

Smithsonian Contributions to Zoology. No. 49: A Revision of North American Epigean Species of *Asellus* (Crustacea: Isopoda). By D. W. Williams. Pp. 80. \$1.25. No. 51: Studies on Ophiocomid Brittlestars. I. A New Genus (*Clarkcoma*) of Ophiocominae with a Reevaluation of the Genus *Ophiocoma*. By Dennis M. Devaney. Pp. 41. \$0.50. No. 57: A Review of the Beetles of the Genus *Metachroma* Chevrolat (Coleoptera: Chrysomelidae). By D. H. Blake. Pp. 109. \$1.50. No. 61: Gammaridean Amphipoda from a Deep-Sea Trench Off Oregon. By J. L. Barnard. Pp. 86. \$1.25. (Washington, DC: Smithsonian Institution Press, 1971.) For sale by US Government Printing Office. [173]

US Department of the Interior: Geological Survey. Professional Paper 658-B: Cretaceous Paleogeography of Southeastern Arizona and Adjacent Areas. By P. T. Hayes. Pp. iii+42. \$0.50. Professional Paper 664: Carbonate Facies and the Lithostrotionid Corals of the Mississippian Kogruk Formation, DeLong Mountains, Northwestern Alaska. By A. K. Armstrong. Pp. vi+38+15 plates. \$1. Professional Paper 680-E: Smaller Foraminifera from Midway Drill Holes. By Ruth Todd and Doris Low. Pp. iii+49+12 plates. \$1. (Washington, DC: Government Printing Office, 1970.) [173]

US Department of Commerce: Environmental Science Services Administration. Collected Reprints—1969. Atlantic Oceanographic and Meteorological Laboratories, Pacific Oceanographic Laboratories Volumes 1 and 2. (Washington, DC: Government Printing Office, 1970.) Vol. 1, \$4.25. Vol. 2, \$3.75. [173]

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Agricultural Science and Human Welfare. By Dr. B. P. Pal. (Presidential address, Fifty-Eighth Indian Science Congress, Bangalore, 1971.) Pp. 10. (Calcutta: Indian Science Congress Association, 1970.) [183]

US Department of the Interior: Geological Survey. Water-Supply Paper 1966: Quality of Surface Waters of the United States, 1965. Parts 12-16: Pacific Slope Basins in Washington and Upper Columbia River Basin to Hawaii and other Pacific Areas. Pp. xiii+492. (Washington, DC: Government Printing Office, 1970.) \$2. [183]

A Checklist of the Birds of Ethiopia. By Emil K. Urban and Leslie H. Brown. Pp. 143. Bibliography of the Avifauna of Ethiopia. By Emil K. Urban. Pp. 28. (Addis Ababa: Haile Selassie I University Press, 1970 and 1971.) [183]

The Paper Industry's Part in Protecting the Environment. Pp. 32. (New York: American Paper Institute, 260 Madison Avenue, 1971.) [183]

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Institut Royal Météorologique de Belgique. Publications Série B, No. 62: Variations Qualitatives et Quantitatives des Composantes du Rayonnement Solaire sur une Surface Horizontale par ciel Serein en Fonction du Trouble Atmosphérique. Par R. Dogniaux. Pp. 22. (Bruxelles: Institut Royal Météorologique de Belgique, 1970.) [193]

US Department of the Interior: Geological Survey. Bulletin 1313-B: Geometric Factors of Bipole-Dipole Arrays. By Adel A. R. Zohdy. Pp. iv+26. (Washington, DC: Government Printing Office, 1970.) \$0.25. [193]

An Approach to the Design of Special Schools for Handicapped Children. By C. A. Urru. Pp. 20. (Pretoria: Council for Science Industrial Research, 1970.) [193]

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